PB-241 810

EVALUATION OF HEALTH HAZARDS ASSOCIATED WITH SOLID WASTE/SEWAGE SLUDGE MIXTURES

William L. Gaby

East Tennessee State University

Prepared for:

National Environmental Research Center

April 1975

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TECHNICAL REPORT DATA
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1. REPORT NO.	2.	PB 241 810
EPA-670/2-75-023		

4. TITLE AND SUBTITLE	5. REPORT DATE
EVALUATION OF HEALTH HAZARDS ASSOCIATED WITH SOLID WASTE/SEWAGE SLUDGE MIXTURES	April 1975; Issuing Date
	6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S)	8. PERFORMING ORGANIZATION REPORT NO.
William L. Gaby	
9. PERFORMING ORGANIZATION NAME AND ADDRESS	10. PROGRAM ELEMENT NO.
Department of Health Sciences East Tennessee State University Johnson City, Tennessee	1DB064;ROAP 24ALU;Task 03
	11. CONTRACT/GRANT NO.
	68-03-0128
12. SPONSORING AGENCY NAME AND ADDRESS	13. TYPE OF REPORT AND PERIOD COVERED
National Environmental Research Center Office of Research and Development U.S. Environmental Protection Agency Cincinnati, Ohio 45268	Final
	14. SPONSORING AGENCY CODE

15. SUPPLEMENTARY NOTES

Contact - Clarence A. Clemons 513/684-4481

16. ABSTRACT

This report summarizes and evaluates the health hazards associated with municipal solid waste/sewage sludge composting by the windrow composting process. The occurrence and survival of pathogens, parasites, and indicator bacteria at various stages during the composting process are described. The study shows that windrow temperatures of 120F to 167F (49C-74C) maintained for at least 7 days destroy pathogens and human parasites. Dog parasitic ova, however, remain intact 35 days after exposure. Considerable variation in the temperature is found at the top and bottom 2 inches of the windrow indicating that proper turning of the compost is essential to ensure destruction of pathogens and parasites. It is concluded that a properly composted solid waste or solid waste/sewage sludge mixture is microbiologically acceptable as a soil conditioner for gardens, farms, and lawns, or for filling areas of erosion without creating health hazards.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Composting Survival Parasites Sewage Sludge	Solid waste Sewage sludge Pathogens Health aspects	13B

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS *(This Report)*	21. NO. OF PAGES
RELEASE TO PUBLIC	UNCLASSIFIED	56
	20. SECURITY CLASS *(This page)*	22. PRICE
	UNCLASSIFIED	4.25

EPA Form 2220-1 (9-73)

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NATIONAL TECHNICAL INFORMATION SERVICE
U.S Department of Commerce
Springfield, VA. 22151

U.S. GOVERNMENT PRINTING OFFICE: 1975-657-592/5359 Region No. 5-11

EPA-670/2-75-023
April 1975

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By

William L. Gaby
Department of Health Sciences
East Tennessee State University
Johnson City, Tennessee

Program Element No. 1DB064

Project Officers

Mirdza L. Peterson and Clarence A. Clemons
Solid and Hazardous Waste Research Laboratory
National Environmental Research Center
Cincinnati, Ohio 45268

NATIONAL ENVIRONMENTAL RESEARCH CENTER
OFFICE OF RESEARCH AND DEVELOPMENT
U.S. ENVIRONMENTAL PROTECTION AGENCY
CINCINNATI, OHIO 45268

For sale by the Superintendent of Documents, U.S. Government Printing Office, Washington, D.C. 20402

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FOREWORD

Man and his environment must be protected from the adverse effects of pesticides, radiation, noise and other forms of pollution, and the unwise management of solid waste. Efforts to protect the environment require a focus that recognizes the interplay between the components of our physical environment--air, water, and land. The National Environmental Research Centers provide this multidisciplinary focus through programs engaged in

- studies on the effects of environmental contaminants on man and the biosphere, and
- a search for ways to prevent contamination and to recycle valuable resources.

This report summarizes and evaluates the health hazards associated with municipal solid waste--sewage sludge composting by the windrow composting process. It is concluded that a properly composted solid waste or solid waste-sewage sludge mixture is microbiologically acceptable for many uses without creating health hazards.

A. W. Breidenbach, Ph.D.
Director
National Environmental
Research Center, Cincinnati

ABSTRACT

The composting of refuse-sewage sludge by the windrow process results in the aerobic biodegradation of organic solids and liquids to a relatively stable end product which may be used as a soil conditioner without creating health hazards or pollution of water, land or air. Composting is a rapid and natural process by which all organic matter is decomposed by microorganisms to inorganic compounds or elements which are utilized by other plants and animals. This cyclic transformation is an essential process without which all plant and animal life would cease. Anaerobic biodegradation also occurs in such processes as landfills but at a much slower rate. The microbial ecology of compost is directly related to the internal temperature of the windrow. These studies indicate that large numbers of microorganism present in refuse and sewage sludge utilize the nutrients available, releasing excessive energy which increases the temperature of the windrow to a maximum of approximately 167 F (74 C) within 7 days. The disappearance of inserted selected pathogenic microorganisms from compost is directly related to this temperature increase and not to any type of antagonistic action resulting from antibiotic activity or other metabolic products of microorganisms in compost. Proper processing, such as aeration and moisture is required for the windrow to reach a temperature of 120 F to 167 F (49 C to 74 C) or greater for a period of 4 to 7 days. If the windrow temperature does not reach 120 F (or falls below 120 F), the microbial flora and pathogens remain viable at a high level and may increase in numbers.

Temperatures observed in the top and bottom 2-4 in layers of the windrows were extremely variable and could not ensure the destruction of pathogens unless the windrows were properly turned.

The handling and disposal of refuse and refuse-sewage sludge should be considered a health hazard and a potential source of many microbial infections.

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CONCLUSIONS

The end product of refuse-sewage sludge composting by the windrow process results in a relatively stable product comprised primarily of humus. These studies have shown that a properly composted refuse or refuse-sludge mixture can safely be used as a soil conditioner for gardens, farms, and lawns or as an ideal material for filling gullies and areas of erosion without creating public health hazards. The disappearance of pathogenic microorganisms from compost was found to be directly related to temperature. Proper composting, therefore was dependent upon the windrow reaching a temperature of 120F-167F (49C-74C) or greater for a period of at least 4 to 7 days. If for any reason, i.e., extreme cold temperatures, improper turning, or anaerobiosis, the windrow temperature does not reach 120 F or falls below 120 F during the process the bacterial flora, including the pathogens originally present in refuse and sewage sludge remains at a high level or even increases significantly in numbers.

The center or midpoint of the windrow reaches a consistently high temperature of 120 F and greater, assuring the destruction of pathogens. However there is considerable variation in the temperature of the top and bottom 2 in. of the windrow indicating that proper turning and mixing of the compost is essential to ensure destruction of all pathogens in windrow composting.

Species of Salmonella and Shigella are present in raw refuse and sewage sludge in relatively small numbers. However these gram-negative, pathogenic enteric bacilli originally present or inserted into the refuse-sewage sludge windrows under controlled conditions, disappeared from the windrow within 7 to 21 days.

Enteroviruses were not isolated from raw refuse, sewage sludge or refuse-sludge compost. Type 2 poliovirus inserted into the windrows were inactivated after 3 to 7 days exposure to 120 F. It is evident from these results that enteroviruses do not present a public health hazard in the properly processed compost and that polioviruses do not survive the composting environment.

Intact parasitic ova could be observed throughout the composting process. However the ova observed in the finished product were not of human origin and would not limit the use of compost. Human parasitic cysts and ova inserted in the center of the windrow were disintegrated after 7 days exposure. Dog parasitic ova remained intact 35 days after insertion in windrows at 2 in. and mid-depth. Knowing that pet excreta may be present in residential refuse the survival of these intact ova should be further investigated. However one would expect to find under normal conditions significant numbers of animal parasitic ova in all suburban areas in which there are pet dog and cat population.

The spirochate Leptospira philadelphia was found to be extremely sensitive to composting temperatures and did not survive for more than 2 days when inserted in windrows.

Several genera of molds where observed in the refuse-sewage sludge compost throughout the process and were especially numerous during the latter stage. The common occurring, easily recognized genera included Mucor, Rhizopus, Penicillium, Aspergillus, Cladosporium and Cephalotecium. The pathogenic fungi Blastomyces dermatitidis and Histoplasma capsulatum were never isolated from raw refuse-sewage sludge mixtures and insertion studies indicated that these pathogenic fungi did not survive composting temperatures. In only one instance did a culture of Histoplasma capsulatum survive for as long as 26 days and this culture has been inserted in the top 2 to 4 in. of the windrow where there was considerable temperature variation. Cultures of Aspergillus fumigatus were readily killed when inserted in the windrows at temperatures of 120 F or greater. However this mold is normally present in refuse and could be isolated from the compost throughout the process.

Insertion studies carried out by Morgan (18) revealed that Mycobacterium tuberculosis (avirulent M. tuberculosis var. hominis $H_{37}RA$) was destroyed within 2 weeks by the high temperatures attained in windrow composting.

Proteus species and in particular, Proteus mirabilis was present in all refuse or raw refuse-sludge compost samples examined. Furthermore, Proteus could consistently be

isolated from soil in which refuse had been mixed. When the refuse decomposed, Proteus could no longer be isolated. These results suggest that Proteus may be an idea indicator organism for the presence of raw or partially decomposed refuse or compost.

The most significant finding of public health interest or concern resulting from this study is that the public health hazards resulting from the handling and disposal of raw refuse are equivalent to or greater than those resulting from the handling and disposal of raw sewage sludge. These results indicate that the total microbial flora of refuse is equal to that of raw sewage sludge. Not only does refuse contain all of the microorganisms found in raw sewage sludge but also all of the organisms found in the upper respiratory tract of man (12). Therefore the handling of and the exposure to raw refuse should be considered a health hazard and a potential source of numerous infections including impetigo, a contagious disease caused by staphylococci and streptococci, diphtheria, streptococcus sore throat, upper respiratory infections including influenza and the common cold.

The disposal of raw solid waste is fundamentally a health problem (1, 24). The results of this study clearly indicate that disposal of raw refuse in open dumps or in landfills constitutes as great a public health hazard as would the dumping of raw dewatered sewage under the same conditions. Covering raw refuse with soil creates an anaerobic condition, comparable to that of an anaerobic windrow, under which sufficient heat is not generated to kill undesirable microorganisms and since most of the pathogenic bacteria found in raw refuse are facultative anaerobes these organisms would survive. Since landfilling is the only method of solid waste disposal recommended by the Environmental Protection Agency (3) the health hazard associated with landfilling should be thoroughly investigated. Such a study has never been made.

RECOMMENDATIONS

1. The most significant finding of public health interest or concern resulting from this study is that the public health hazards resulting from the handling and disposal of raw refuse are equivalent to or greater than those resulting from the handling and disposal of raw sewage sludge. These results indicate that the total microbial flora of refuse is equal to that of raw sewage sludge.

2. Based on these studies aerobic windrow compost where properly processed, does not constitute a public health hazard and is an excellent soil conditioner for gardens, lawns and other areas. Compost should not be limited in its use any more than any other soil conditioner.

3. Use of material from anaerobic windrows would constitute a public health problem since microorganisms are not destroyed.

ADDITIONAL RESEARCH NEEDS

1. An educational program should be initiated to inform the general public of health hazards associated with the handling of raw solid waste in the home and recommend types of refuse disposal containers.

2. Additional studies should be made to confirm the results indicated in this study of the possible thermo resistance of dog parasitic ova.

3. Studies are needed to determine possible means of reducing the cost of recycling solid wastes.

4. Research is needed to determine the possible use of 14 to 21 day old compost for various reclamation projects.

5. Reduce cost of recycling by homeowners separation of biodegradable and nonbiodegradable wastes through local ordinance.

6. Since the only method of solid waste disposal recommended by the Environmental Protection Agency is landfilling, more intensive studies should be made to determine the survival of microorganisms in solid waste under these anaerobic conditions.

INTRODUCTION

This report summarizes and evaluates the health hazards associated with solid waste sewage sludge composting from data accumulated during the period from June 1, 1967 through May 31, 1969 under contracts No.PH-86-67-112 and No.86-68-143 between the U.S. Public Health Service and East Tennessee State University, Department of Health Sciences, College of Health, Johnson City, Tennessee. The study was initiated as a result of a cooperative joint project agreement between the U.S. Public Health Service, The Tennessee Valley Authority and the Municipality of Johnson City, Tennessee (15).

The scope of work included in these contracts was divided into two parts.

Phase 1 included the development of methods to determine the presence of a) total coliforms (MPN), b) fecal coliforms (MPN), c) Salmonellae, d) Shigellae, e) coagulase positive staphylococci, f) Protozoa (Entamoeba histolytica and Entamoeba coli), g) Ova of Cestodes (Taenia sp., Diphyllibotrium latum, Diphylidium caninum, Echinococcus sp.) h) Ova of Nematodes (Ascaris lumbricoides, Strongyloides sp., Enterobius vermicularis, Trichuris trichiura), i) Histoplasma capsulatum, j) Candida albicans, k) Aspergillus fumigatus, and l) Enteric viruses (ECHO, Cocksackie, and Polio).

Phase 2 included the development of methods to determine the survival patterns of and the percentage reduction of the following organisms inserted in the windrows by the thermophysical processes or by antagonistic action caused by microbial competition or by antibiotic inhibitors: a) bacteria: Escherichia coli, Salmonella typhimurium, Shigella sonnei, Staphylococcus aureus (coagulase-positive), b) parasites; Entamoeba histolytica, Ascaris lumbricoides (viable ova); c) fungi: Histoplasma capsulatum Aspergillus fumigatus, Geotrichum candidum; d) viruses: Poliovirus, e) spirochaeta: Leptospira philadelphia.

The aerobic biodegradation of organic solids and liquids to a relatively stable end product comprised primarily of humus is a satisfactory method for the disposal of solid waste without creating health hazards and water, land or air pollution (16). Since the finished product is an excellent soil condi-

tioner, (17, 20, 26) aerobic thermophilic decomposition of urban, refuse and sewage sludge may be applied for recycling of municipal waste.

Debate over the use of compost as a soil conditioner varies from those who advocate its usefulness and safety (4, 5, 8, 13, 16, 17, 19, 20, 22, 23, 26) to those who question its usefulness or safety (9, 21). However, it has been generally assumed that a correctly managed composting process is adequate in destroying the diverse flora of pathogenic microorganisms present in such an environment. Previous studies have indicated that microorganisms are killed as a result of the high temperatures obtained during composting. However some investigators have indicated that an antibiotic action is responsible for destruction of some pathogens (26). Knoll (16) reported killing of Salmonella species within 2 to 7 days at compost temperatures of 122 F (50 C). At temperatures below 112 F (45 C) destruction of all Salmonella strains cannot be assured. In a later report Knoll (17) found that typhoid bacteria were destroyed in compost at a temperature of 112 F after 7 to 9 days and in ampules up to 257 days and that these bacteria can resist the complex antagonistic processes in composting material for 247 days. Niese (19) concluded from his studies that the self-heating of compost is dependent upon the availability of nutrients and not upon the numbers of microorganisms present, but the speed at which the maximum temperature is reached is influenced by the total number of microorganisms present. Farkasdi (10) reported that rapid environmental changes initially enhance the development of large numbers and varying species of microorganisms followed by a heating period necessary for the destruction of pathogenic organisms. He also indicated that the presence of fungi in compost is related to its moisture content. The drier the compost the more numerous the fungi. The mesophilic fungi are numerous at 98 F (37 C), numerous thermophilic fungi at higher temperatures and a complete absence of fungi at 150 F (67 C).

Rohde (22) found that Ascaris ova were destroyed after 4 days in decomposing slaughter house wastes. He reported that Ascaris ova are killed in 10 days at 104 F (40 C) and within 24 hr at 140 F (60 C). Scott (23) reported that Endamoeba histolytica, Endamoeba coli, and Ascaris ova are destroyed within three weeks in windrow composting. Strauch (25) reports that the psittacosis virus is inactivated when exposed

to the environmental conditions in composting that are lethal to pathogenic bacteria.

Studies by Sliepcevich (24) indicated that the handling and disposal of raw solid wastes constitutes an improper health hazard. She found from a study of the Department of Sanitation of New York City that arthritis, cardiovascular diseases, muscle and tendon diseases, and skin diseases could be classified as occupational diseases of refuse collectors. Anderson (1) also stated that the disposal of solid waste is fundamentally a health problem.

MATERIALS AND METHODS

The Johnson City open windrow composting plant processed city refuse from which as much of the nonbiodegradable materials as possible were removed (15). The refuse was ground into small particles of from 1 to 3 in. in diameter, mixed with 3 to 5% primary dewatered sewage sludge and deposited by dump truck in windrows approximately 4 to 5 ft high, 7 to 10 ft wide and 150 ft long. The windrows were turned mechanically at specified intervals of several days to insure proper aeration and thorough mixing. The moisture content was maintained at 50 to 60%. The composting process was usually completed within 49 days.

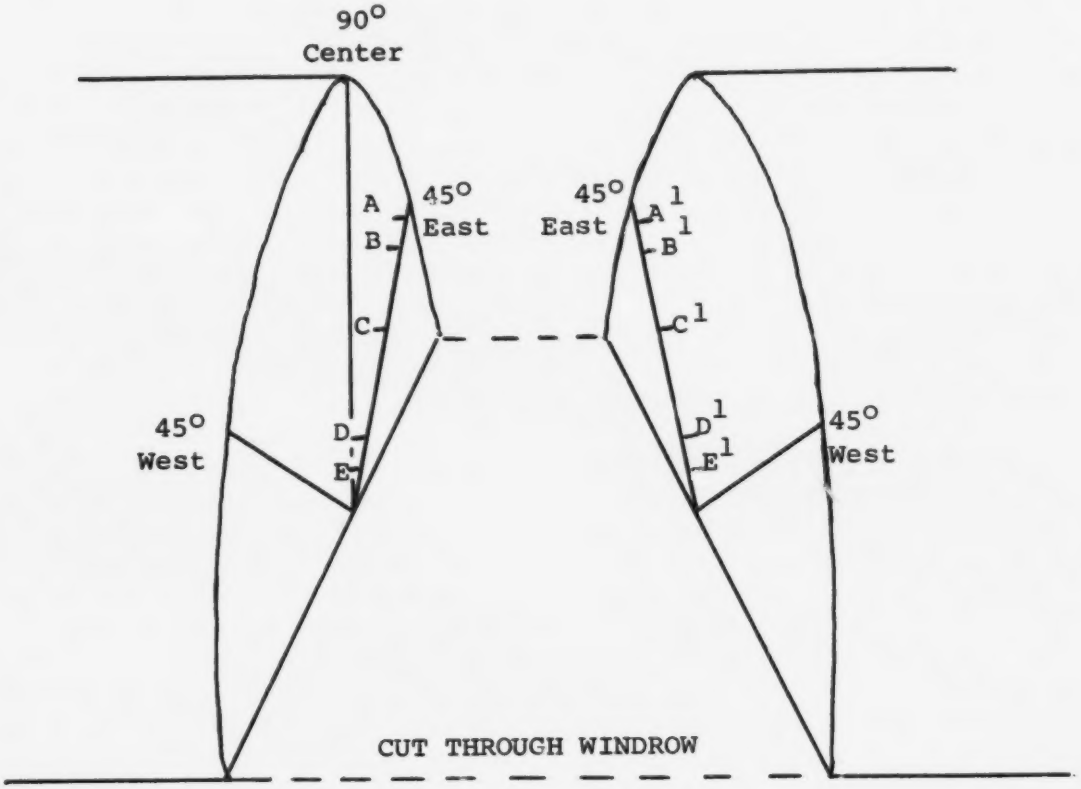
A variety of specimens, including raw refuse, raw sewage, digested sludge and sewage sludge-refuse from the windrow area, were received from the Johnson City plant. U. S. Public Health Service personnel were responsible for collecting all specimens, including the methods for collecting samples, the type of samples and the number of samples obtained from each windrow. Figure 1 illustrates a typical cross section of the windrow and the locations from which samples of compost were obtained for examination.

Sampling procedures. A windrow was cut through from top to bottom to expose two cross sections. Samples were taken from each section of the windrow at the locations shown in Figure 1. Samples were collected with tongs which were sterilized in the field between each use. Certain samples were combined such as A with A^1; B with B^1; etc. Samples were placed in sterile plastic bags for transportation.

To insure uniformity and compatibility of all data, dry weights of all specimens were determined and the results are expressed as the number of microorganisms per g dry weight.

Dry weights were determined by placing a sample of known weight in a vacuum oven at 176 F (80 C) for 2 to 5 days until all of the moisture was removed as indicated by the consistent weight of the sample. The percent moisture of each specimen was calculated from the dry weights.

Due to the heterogeneous nature of compost, preliminary experiments were carried out to determine the most accurate



TYPICAL CROSS-SECTIONS

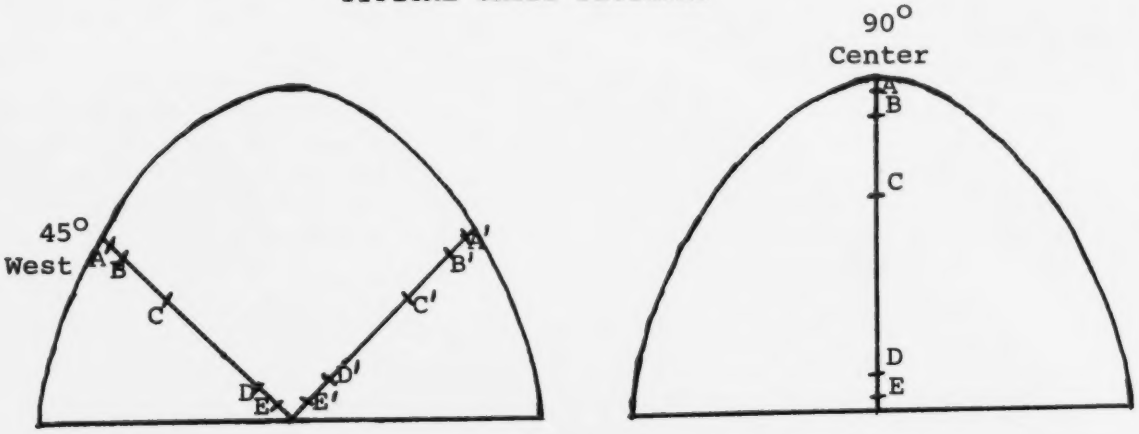


FIG. 1. Cross sections and sampling locations. A = 2-4 in. below surface, B = 6-8 in. below surface, C = 2 ft below surface, D = 6 in. above bottom, E = 2 in. above bottom.

method for preparing a homogeneous aqueous suspension of the specimens in order to get consistent results. Approximately 70 to 100 g random samples of compost were obtained in plastic bags. After thoroughly mixing, 5 g sub-samples were suspended in 95 ml of saline (0.85% salt solution), 0.1 M phosphate buffer at pH 7.0, and distilled water. Suspension of the compost sample in each of these diluents were effected by: 1) vigorous manual shaking in a standard 100 ml dilution bottle; 2) mechanical shaking on a rotary shaking machine describing a circle one in. in diameter at 200 rpm for a period of one min; 3) homogenizing in an Oster blender set at slow speed (stir) for one min. Triplicate plate counts were made on each suspension.

Of the three diluents tested, saline and 0.1 M phosphate buffer gave consistently higher and more reproducible results than did distilled water. It was decided therefore to use saline as the standard diluent because of the large quantities required, simplicity in preparation and less chance of error. For suspending the compost in the diluent, homogenizing gave consistent higher and more reproducible counts than did manual or mechanical shaking and was therefore adopted as the standard means for suspending compost samples throughout the study.

Microbiological procedures

1. Total and fecal coliforms: Coliform counts were determined by suspending 5 g weight samples of compost in 95 ml of sterile saline (1-20 dilution). Duplicate 5 g samples were also taken for dry weights determinations. Each suspension was homogenized for 1 min, and processed according to procedures described in Standard Methods for the Examination of Water and Wastewater (2) by the use of the 5 tube technique for detecting and quantitating total and fecal coliform densities. All results are expressed as the number of organisms per g dry weight of sample.

2. Fecal streptococci: The presence of fecal streptococci was determined by inoculating aliquots of the serially diluted compost suspensions into duplicate tubes of KF Streptococcal broth. This procedure was selected as being more conducive than the KF Streptococcal agar when the two media tested simultaneously gave compatible results. All tubes of KF Streptococcal broth were checked for the presence of growth after 48 hr incubation at 95 F (35 C). Periodic examinations

of the growth in these tubes were made to confirm the presence of streptococci.

3. Coagulase positive staphylococci: In selecting a medium for the isolation and identification of Staphylococcus from compost, Chapman-Stone, TPEY, Staph 110 and 5% blood agar were compared. We found that colonies of Staphylococcus isolated from compost were more easily recognized on Staph 110 than they were on the other types of media. Appropriate dilutions (10^2, 10^4, and 10^6) of the compost suspensions were plated on Staph 110 medium, incubated at 98.6 F (37 C) for 24 to 48 hr and the small, round, glistening, entire, yellow to golden pigmented colonies were subcultured and subsequently tested for coagulase activity.

4. Salmonella and Shigella: For the detection of Salmonella and Shigella species direct plating proved to be unsatisfactory, since a prohibitive amount of overgrowth was exhibited. Dilutions of the compost suspension, in an attempt to overcome excessive growth, resulted in uniformly negative results. Therefore, selective enrichment media were used: Selenite F enrichment broth and Selenite brillant green enrichment broth containing sulfapyridine (SBG sulfa). Of these two media, the SBG sulfa enrichment was more selective. Thirty g of the compost samples (undiluted) were placedinto 270 ml of enrichment media and incubated at 106 F (41.5 C) in a controlled temperature water bath for 18 to 24 hr. After incubation several loopsful of the enrichment broth culture were streaked, in triplicate, on Salmonella-Shigella (SS) agar, bismuth sulfite (BS) agar and MacConkey agar respectively. All plates were incubated at 95 F (35 C) for 18 to 24 hr and checked for typical and suspected colonies of Salmonella and Shigella. Suspected colonies were transferred to triple sugar iron (TSI) agar slants, incubated at 95 F for 24 hr and examined for typical reactions indicative of Salmonella and/or Shigella species (7). Suspected positive cultures from TSI agar were inoculated into urea broth, lactose broth, and semi-solid agar. All cultures whose biochemical reactions were typical of Salmonella or Shigella species were submitted to the Tennessee Department of Public Health Laboratories, Nashville, Tennessee for final serological identification by standard procedures recommended by the Center for Disease Control.

5. Total plate count: Five grams of compost were suspended in 95 ml of sterile saline and homogenized at lowest speed for 1 min. From this initial suspension, dilutions of 10^2 through 10^8 were inoculated into sterile petri plates in replicates of eight. Since preliminary experiments had shown comparable results using trypticase soy agar (TSA) and Standard methods agar (SMA) as plating media, TSA was chosen as the media to be routinely used. These plates were incubated at 95 F (35 C) and at 131 F (55 C) both aerobically and anaerobically for 48 hr to determine the total number of colonies. Anaerobic conditions were achieved via Gas Pak Anaerobic jars.

6. Enteroviruses: Approximately 20 to 30 g of each sample received from the Johnson City Composting Plant were frozen in sterile plastic bags, packed in dry ice, and shipped air mail to Tennessee Department of Public Health Laboraotries in Nashville. Upon receipt of the frozen specimens in the State Laboratory, each sample was allowed to thaw completely at room temperature, and the contents of the same bag were mixed thoroughly by manipulation of the bag. Approximately 2 g of the sample were removed and placed in a flask containing 20 ml of cold, sterile distilled water and glass beads. Preliminary examinations indicated a 2 g sample was adequate. The flask was shaken vigorously to mix the contents and the suspension was poured into a sterile centrifuge tube. The suspension was then clarified by centrifugation in a refrigerated centrifuge (4 C) at 1500 rpm for 20 min. The supernate was poured off and re-centrifuged for 1 hr at 3000 rpm. The clear supernate was removed from the sediment and an antibiotic solution was added to give a final concentration per ml of 1000 units of penicillin and 1000 ug of streptomycin. The sample was held at room temperature for 30 min. The sample was then inoculated into 3 tubes of primary monkey kidney cells (African Green purchased from Microbiological Associates) and 3 tubes of Hep 2 cells (maintained by serial passage in the State Laboratory). The tubes were incubated on a roller drum at 98.6 F (37 C) for 8 to 9 days. All cell cultures were observed daily for virus activity (6).

7. Parasites: Three methods were employed in the parasitological examination of the compost samples: 1) direct mount; 2) brine flotation; and 3) formalin-ether sedimentation. To prepare the samples for examination, 2 g of compost were placed in a 250 ml flask containing 20 to 30 ml of saline and

enough glass beads to cover the bottom of the flask. After thoroughly shaking to emulsify the sample, the flask was tilted and left undisturbed for at least 30 min to allow sedimentation.

Approximately 0.05 ml of the sediment was placed upon a clean slide for direct microscopic examination (direct mount) Iodine was employed to obtain characteristic differentation.

Following this procedure, the original suspension was thoroughly mixed and strained through a wire mesh funnel into a 50 ml centrifuge tube. The screen was washed with saline and the final volume in the centrifuge tube was brought to approximately 45 ml. After centrifugation at 2000 rpm for 1 to 2 min, the supernatant was decanted and the sediment resuspended in 20 ml of saline. Ten ml of this suspension was removed and saved for formalin-ether sedimentation proceudre. The remaining 10 ml was centrifuged at 2000 rpm for 1 to 2 min and decanted. The sediment was resuspended in approximately 20 ml of brine (1.2 sp. gr.) and transferred to a shell vial. The vial was filled to the lip with brine and a clean glass slide was carefully superimposed over the vial, taking care to avoid overflow and air pockets. This preparation was allowed to stand 10 to 15 min and the slide was carefully removed and fitted with a cover glass. The edges were sealed with Vaspar and microscopic examination was made. This constituted the brine flotation method.

The remaining 10 ml of suspension was centrifuged at 2000 rpm for 1 to 2 min, and the supernatant decanted. Approximately 10 ml of 10% formalin was added and after thorough mixing, the suspension was allowed to stand for 5 min. Three ml of ether was added and the tube was shaken vigorously for 30 sec and centrifuged at 1500 rpm for 1 to 2 min. Four layers resulted from this centrifugation; a small amount of sediment containing the ova and parasites, a layer of formalin, a layer of debris, and a layer of ether. The top three layers were carefully decanted and a sample from the bottom layer was used to prepare an iodine mount for microscopic examination.

8. Pathogenic fungi: Five grams of compost samples were added to 100 ml of sterile physiological saline. After shaking to suspend the sample, the supernatant was separated

and centrifuged at 2500 rpm for 15 min. The supernatant was decanted and the sediment was thoroughly mixed and added to sterile screw cap vials containing 10,000 units of penicillin and 10 mg of streptomycin. This suspension was allowed to stand at room temperature for 20 min.

Each of three white Swiss mice (4 to 6 weeks of age) were inoculated intraperitoneally with 0.5 ml of the concentrated sediment. At the end of 3 weeks, the mice were sacrificed and a portion of the liver and the entire spleen were removed and placed in a sterile petri dish. After these tissues were minced, small portions were used to inoculate two tubes of Sabouraud's agar and two tubes of Sabouraud's agar containing 0.5 mg of Actidione (cycloheximide) per ml and 0.05 mg of chloromycetin per liter. All cultures were incubated for 4 weeks, with weekly examinations being made. Smears of suspicious colonies were made and fungi were identified by cultural characteristics.

Two tubes of Sabouraud's agar and two tubes of Sabouraud's agar containing 0.5 mg of Actidione per ml and 0.05 g of chloromycetin per liter was inoculated with a small portion of the concentrated sediment. All tubes were incubated at 25 C, and examined weekly. At the end of 6 weeks, smears of suspicious colonies were made and identified by cultural characteristics (6).

Insertion studies

1. Bacteria: Insertion studies were carried out using culture tubes of *Salmonella typhimurium*, *E. coli*, coagulase positive *Staphylococcus aureus*, and *Shigella sonnei* to determine their survival time in refuse-sludge compost windrows. To study the effect of temperature, as well as the possible effect of an antibiotic activity that might be involved in the disappearance of pathogenic and indicator organisms from compost, the test organisms were placed in closed containers as well as in open contact with the compost.

Three methods were used to study the effect of temperature on the inserted bacteria:

a. Young, 2-12 hr agar slant cultures in 10X75mm cotton plugged test tubes were inserted at various depths for varying periods of time in the windrows.
b. Young, 18-hr nutrient broth cultures in 16X125mm screw cap tubes were also inserted in the windrows.

c. Young, 18-hr nutrient broth cultures were sealed in ampules and inserted into the windrows.

Three methods were used to study the possible effect of antibiotics which might be present in compost.

a. One ml of 18-hr nutrient broth cultures were injected into compost enclosed in small nylon bags.
b. One-to-two ml of 18-hr nutrient broth cultures were placed on sterile cotton balls and inserted directly into the compost.
c. Sterile filter paper discs were saturated with young, 18-hr broth cultures of the organism and placed directly into compost.

To facilitate the recovery of samples inserted into the windrows, small nylon bags were used. Freshly prepared 0 day compost was placed in the bags along with the culture tube (slants, screw cap tubes or ampules). The cotton balls or filter paper discs were placed in the bags in direct contact with compost. The bags were securely tied and inserted into the windrows. Sufficient quantities of these "bagged" samples were prepared to allow four samples to be taken from each depth (2-4 in., midpoint, or bottom) within the sample period.

Recovery of organisms from broth and agar slant cultures: Standard plate counts were run on all broth cultures recovered from the windrows. Agar slant cultures recovered from the windrows were scraped with sterile inoculating loops and streaked on nutrient agar plates.

Recovery of organisms from filter paper discs: *E. coli*, and *Staphylococcus aureus* were recovered from the compost sample, placed in 100 ml of sterile saline, shaken vigorously by hand for one min and 0.1 ml amounts of the saline suspensions were inoculated directly onto duplicate plates of EMB and Staph 110 agar. Following 24 hr incubation, the plates were examined for typical colonies, which were further confirmed by biochemical tests (7).

Salmonella typhimurium: Filter paper discs recovered from the windrows were placed in 100-200 ml SBG sulfa medium and incubated at 106 F (41.5 C) for 18-24 hr. Duplicate plates of SS agar and bismuth sulfite agar were streaked from the

SBG sulfa and examined for typical colonies after 48 hr. Those cultures which warranted further examinations were inoculated into SIM agar, urea broth, and lactose broth. If the results from these indicated the culture to be a Salmonella the culture was sent to the Tennessee Department of Public Health, Nashville, for serological typing.

Shigella sonnei: The filter paper discs were recovered from the windrow, placed in 100-200 ml of Selenite F broth, incubation at 95 F (35 C) for 18-24 hr, and the procedures used for the isolation and identification of Salmonella typhimurium were carried out.

2. Enteroviruses. Suspensions of poliovirus (Type 2) were received from the Tennessee Department of Public Health in a frozen state. Portions of the liquid suspensions were added to screw cap tubes as well as used to saturate filter paper discs. The tubes and discs were placed in nylon bags containing 0 day compost and inserted at all three depths in the windrows. At various time periods, sample nylon bags were removed from the windrows, frozen in dry ice and mailed to the Tennessee Department of Public Health in Nashville for viability studies.

3. Leptospira: Leptospira philadelphia, obtained from Women's Medical College in Philadelphia, Pa. was grown in Fletcher medium base containing 10% leptospira enrichment (Difco). Young cultures in screw cap tubes were inserted into the windrows for various periods of time from 48 hrs to 3 weeks. Upon removal of the culture tubes from the windrows, viability of the organisms was determined by making sub-cultures to fresh medium and observing the growth under a darkfield microscope for motility. Filter paper discs recovered from the windrows were placed in 16mm test tubes containing 3 ml of Fletcher medium base, shaken manually for approximately one minute and the supernatant examined under the darkfield microscope for motile leptospira.

4. Fungi. Young sub-cultures of Histoplasma capsulatum, Blastomyces dermatitidis, Geotrichum candidum, and Aspergillus fumigatus were made on potato dextrose agar or Sabouraud agar slants in 16x125mm screw cap tubes and inserted at various depths into the windrows. At various

periods of time, these cultures were removed from the windrows and sent directly to the Tennessee Department of Public Health, Nashville, for positive identification and confirmation of viability.

5. Parasites. Positive stool specimens were obtained from area hospitals. One positive dog fecal speciman was also included in the study. Each of the stool specimens were divided into several equal samples in plastic bags and inserted in the windrow for varying periods of time. Upon removal from the windrows, the samples were examined by the Tennessee Department of Public Health, Johnson City branch laboratory. Formalin-ether and salt flotation methods were carried out to determine the presence (or absence) of ova in the sample. Viability of the ova was not determined by animal feeding.

Background studies

To better understand the bacterial ecology of composting a study was carried out by two microbiology majors at East Tennessee State University to examine the bacterial flora of compost throughout the process and attempt to find an organism more indicative of compost in the early stages of decomposition than are the coliforms (11). These studies were carried out by taking large samples of compost (50-100 g) at 2 day intervals from 8-16 in. below the surface at approximately midpoint in the windrow. A thoroughly mixed 5 g sample was suspended in 45 ml saline, homogenized for 1 min at slow speed, dilutions prepared in saline from 10^1 through 10^9 and 0.1 ml of each dilution in duplicate was spread evenly over the surface of SS, eosin-methylene-blue, trypticase soy, and Staphylococcus 110 agar plates. Duplicate plates were incubated at 95 F (35 C) and 131 F (55 C) under aerobic and anaerobic conditions for 24 to 36 hr. The resulting colonies were observed macroscopically and further identified by inoculating into appropriate biochemical media, ie., TSI agar, SIM agar, citrate agar, urea medium, and other differential media when required (7).

For comparative studies soil samples were obtained from cultivated fields, pasture land and woodland areas. Five g of each soil sample were examined for the presence of total and fecal coliforms as well as for the presence of Proteus. (12)

RESULTS

The average number of total coliforms, fecal coliforms and fecal streptococci found in raw refuse and sewage sludge are shown in Figure 2. It is of importance to note that the coliform count of raw refuse is equivalent to that of sewage sludge and that the fecal streptococci count of raw refuse is substantially greater than that of sewage sludge.

Season variations in the coliform counts of the windrows was not observed. Although the results shown in Figure 3 would indicate that the coliform counts were somewhat less during the Fall quarter of the year it should be pointed out that during this period of time the project was just getting underway, sampling procedures had not been established, procedures for handling of the raw materials at the composting plant were being investigated, and laboratory procedures had not been standarized. Additional data that could not be included in these average counts indicate that the coliform counts remained fairly constant during the Fall, Winter, Spring, and Summer seasons. Fecal streptococci were made for the Fall and Winter seasons only.

Species of Salmonella were isolated occasionally from raw refuse and sewage sludge only after 20 to 30 g of the sample was cultured in an enrichment medium. Only on one occasion was a Shigella isolated from sewage sludge. This was early in the Fall of 1967 when the project was starting and the species was not identified. The species of Salmonella isolated are listed in Table 1. In no instance was Salmonella found in a windrow after 7 days.

The bacterial ecology of compost was found to be directly related to the internal temperature of the windrow. The freshly ground compost contains large numbers of microorganisms which utilize the carbohydrates present in refuse as indicated by a characteristic drop in pH during the first few days of the composting process. As a result of the release of this excess energy the temperature of the windrow reaches 120 F (49C) to a maximum of 167 F (75 C) within 7 days and remains high throughout the process. If this temperature is not maintained due to anaerobiosis, the bacterial population, (especially the coliforms) remains at a high level or increases in number. Figures 4,

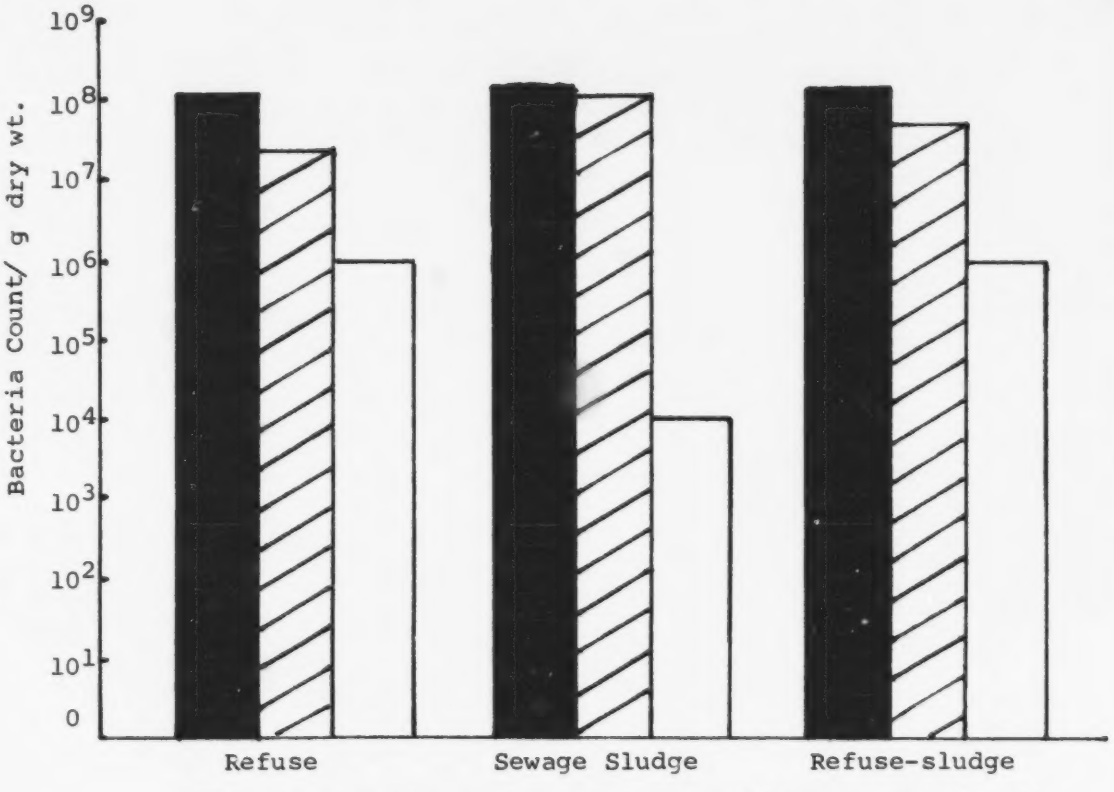


FIG. 2. Average number of total coliforms, fecal coliforms and fecal streptococci in raw refuse, sewage sludge and raw refuse-sewage sludge mixture on zero day. Numbers represented averages of 6 to 8 specimens. Total coliforms ■ ; fecal coliforms ▨ ; fecal streptococci □

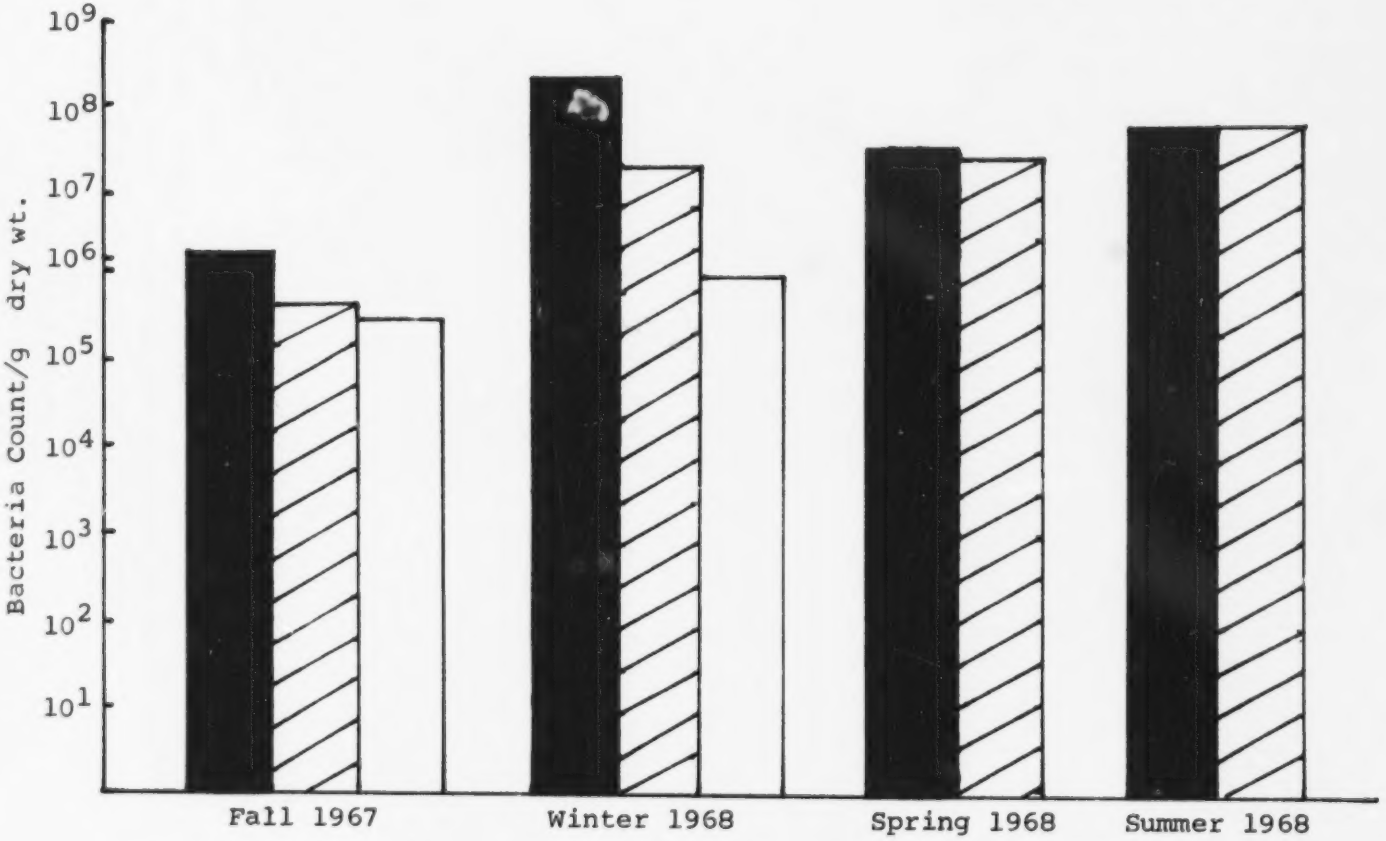


FIG. 3. Average number of total coliforms, fecal coliforms and fecal streptococci in refuse-sewage sludge on zero day for the four seasons. Data of fecal streptococci for Spring and Summer not available. Total coliforms ■ ; fecal coliforms ▨ ; fecal streptococci □

TABLE 1. Isolation of Salmonella from raw-refuse, raw-sewage sludge and refuse-sewage sludge mixtures on 0 day

Raw-Refuse Only	Raw-Refuse Sludge Only	Refuse-Sewage Sludge Only
S. enteritidis	S. enteritidis	S. enteritidis
S. typhimurium	S. typhimurium	S. typhimurium
S. saint paul	S. saint paul	S. heidelberg
S. heidelberg*	S. anatum**	S. saint paul
S. montevideo*	S. chester**	S. braenderup***
	S. derby**	
	S. eimsfuettel**	
	S. muenchen**	

*Isolated only from raw refuse, never found in raw sewage sludge.

**Isolated from raw sewage sludge, never found in raw refuse.

***Isolated on one occasion from refuse-sewage sludge mixture. It was not found in raw refuse or sewage sludge.

Salmonella were isolated from only a small percent of the raw refuse or raw sewage sludge specimens examined.

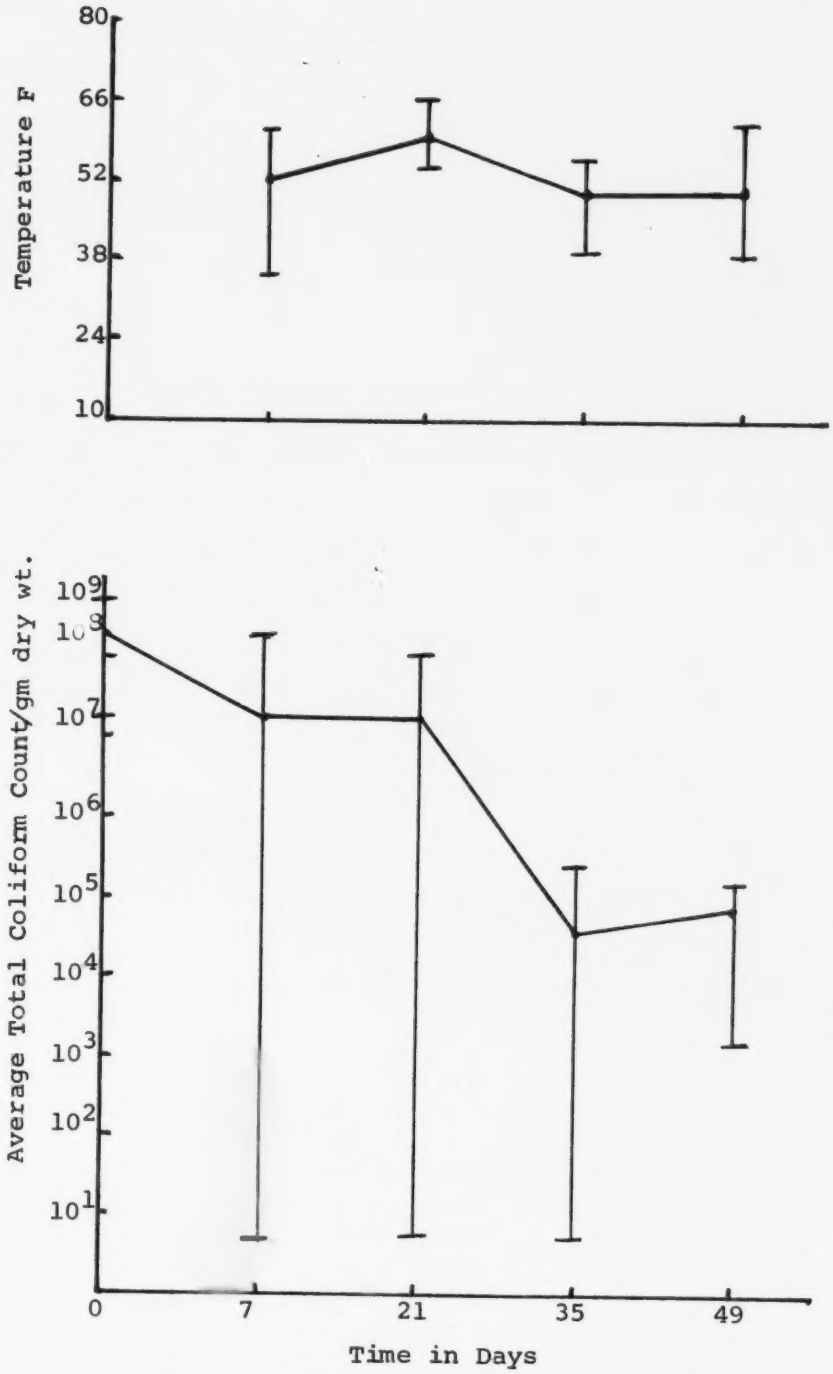


FIG. 4. Average total coliform MPN from eight refuse-sewage sludge windrows. Vertical bars represent extent of variations found in the windrows. 2-4 in. depth.

5, 6, 7, and 8 show the average total coliform counts present throughout the 49 day refuse-sewage sludge compost process. These counts are averages of the results obtained from 8 windrows in an attempt to demonstrate the extent of variations observed between windrows and the various areas within the windrow from which samples were obtained. These results typify the extent of variation observed in samples obtained 2-4 in. from surface (FIG. 4) and 2 in. from bottom (FIG. 8).

The total microbial population in raw refuse and refuse sludge windrow remain relatively constant throughout the composting process as indicated by Figures 9 and 10. There is however a complete change in the windrow flora. As the temperature of the windrows increases the mesophilic bacterial population is rapidly replaced with pseudothermophilic and thermophilic bacteria, maintaining a remarkably constant total bacterial count. The aerobic and anaerobic bacteria population capable of growing at 95 F (35 C) increased in number from 10^4-10^5 on 0 day to approximately 10^8 by the 49th day per g dry weight of compost.

The results of a more detailed study of the bacterial ecology of compost are shown in Figures 11, 12, and 13. It is evident from the results illustrated in Figure 11 that although the coliforms disappeared from compost within the first 2 weeks, they did reappear at various times throughout the process period. This reappearance may be due to recontamination with coliforms from the composting plant equipment, flies and birds, or to the release of organisms from organics during the decomposition process. The fecal streptococci follow the same general pattern as the coliform, but appear to be more resistant to the windrow temperatures than did the coliform.

The mesophilic Bacillus species cannot survive the windrow temperatures and disappear from the compost within two weeks as shown in Figure 12. However, the pseudothermophilic and thermophilic Bacillus species increase in number after the first 2 weeks and remain throughout the process. Numerous colonies of yellow-orange chromogenic bacteria appear early in the process but disappeared rapidly as the windrow temperature increased. These chromogenic bacteria were short gram negative, gelatinase-producing bacilli belonging to the genus Serratia. During the second week

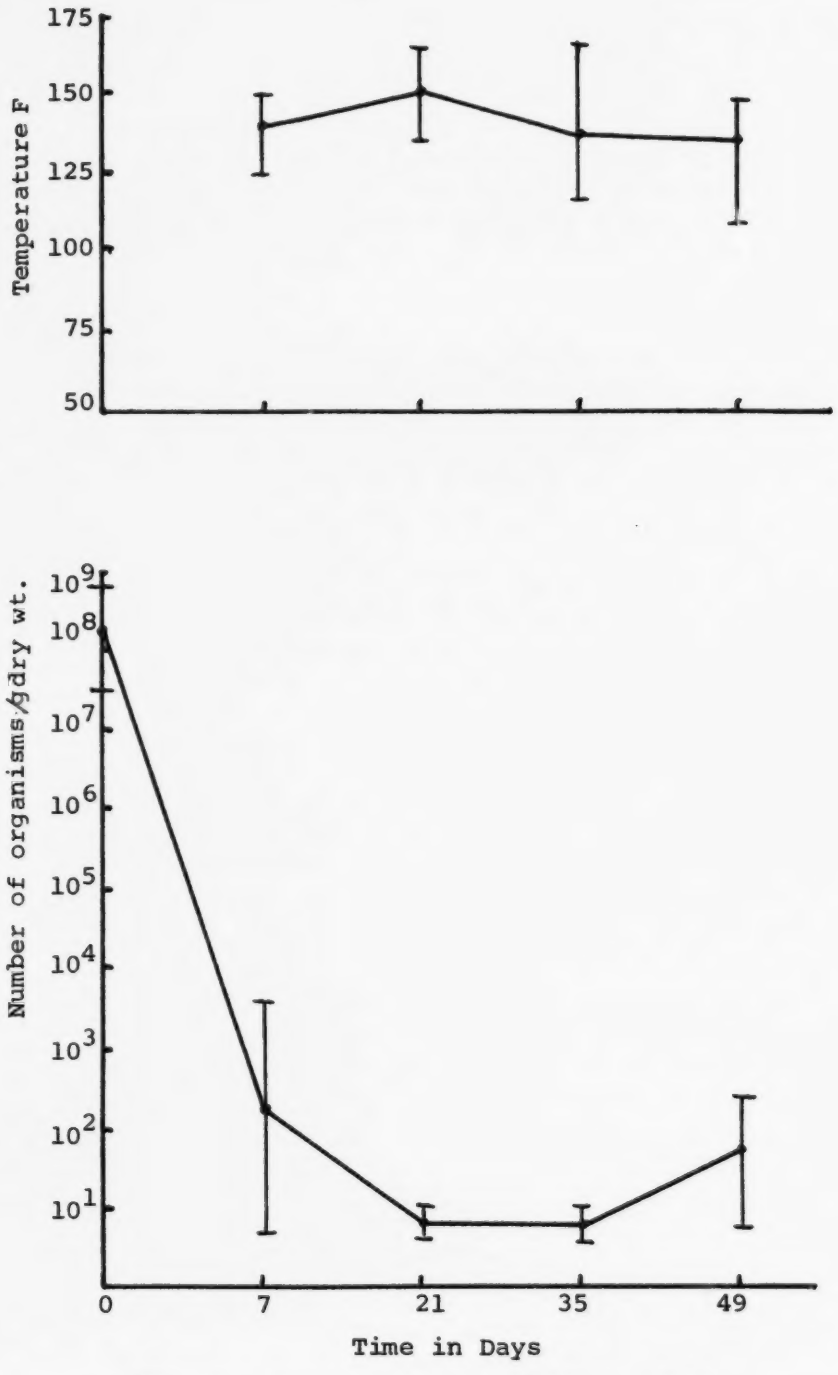


FIG. 5. Average total coliform MPN from eight refuse-sewage sludge windrows. Vertical bars represent extreme variations found in the windrows. 6-8 in. depth.

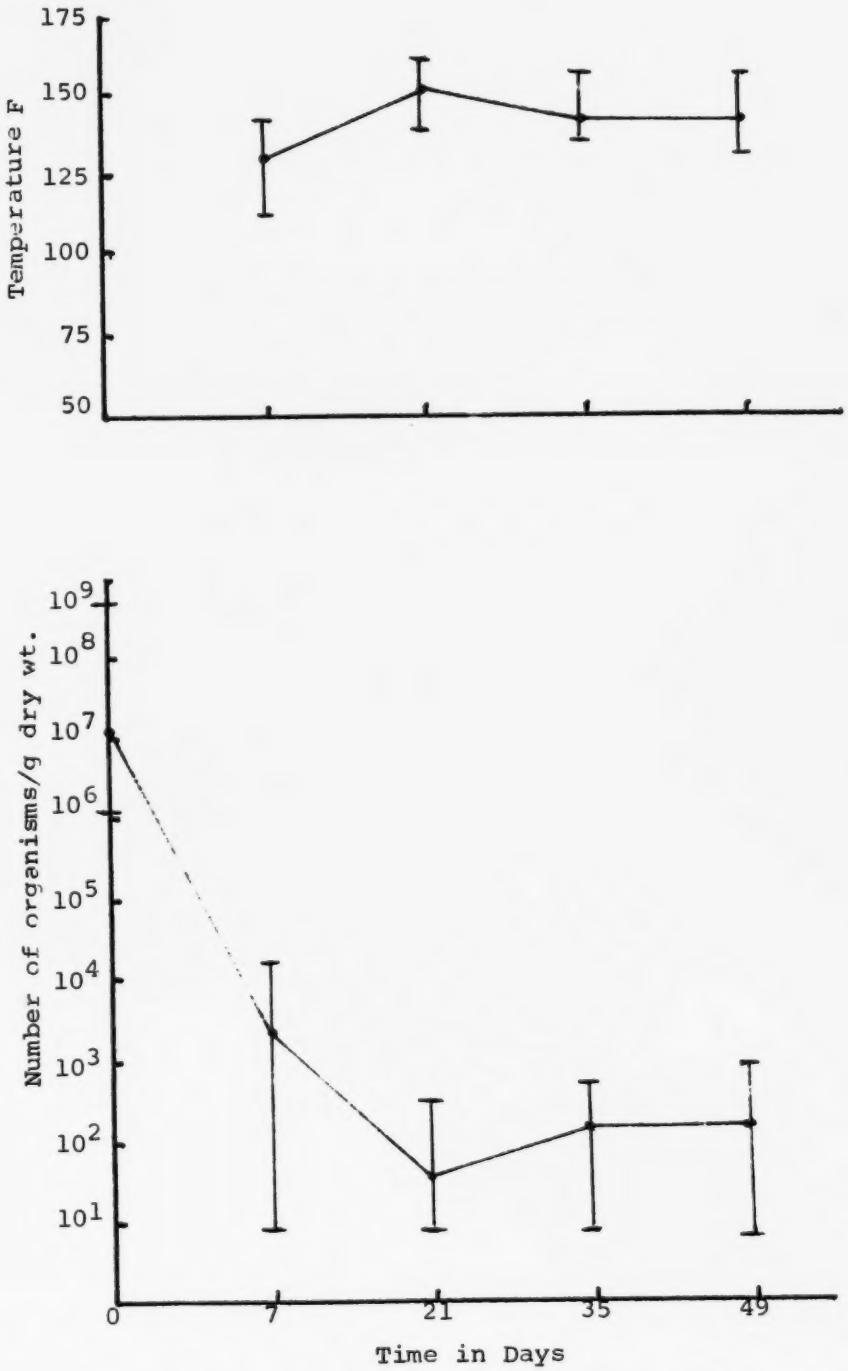


FIG. 6. Average total coliform MPN from eight refuse-sewage sludge windrows. Vertical bars represent extreme variations found in the windrows. 2 ft depth.

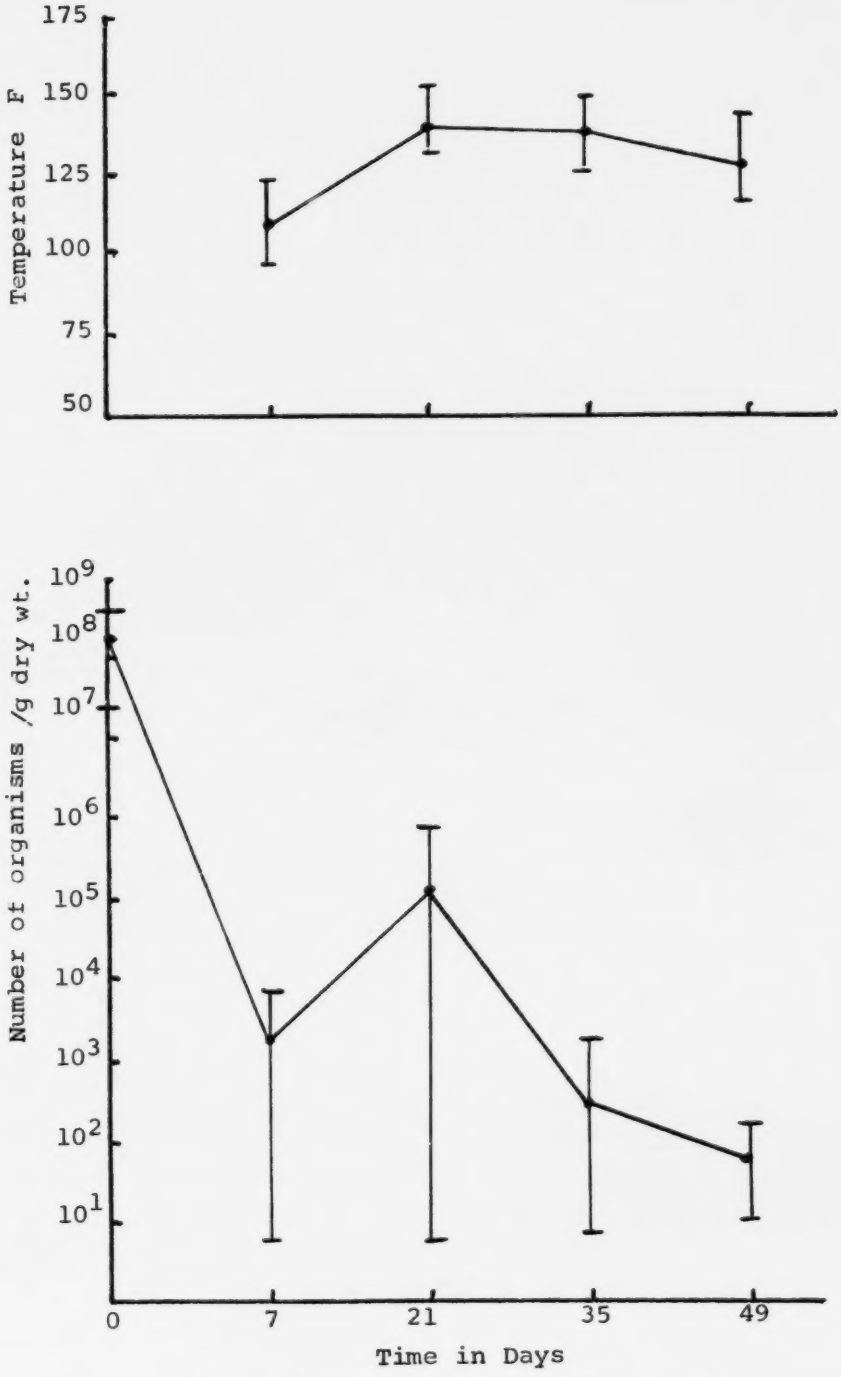


FIG. 7. Average total coliform MPN from eight refuse-sewage sludge windrows. Vertical bars represent extreme variations found in the windrows. 6-8 in. from bottom.

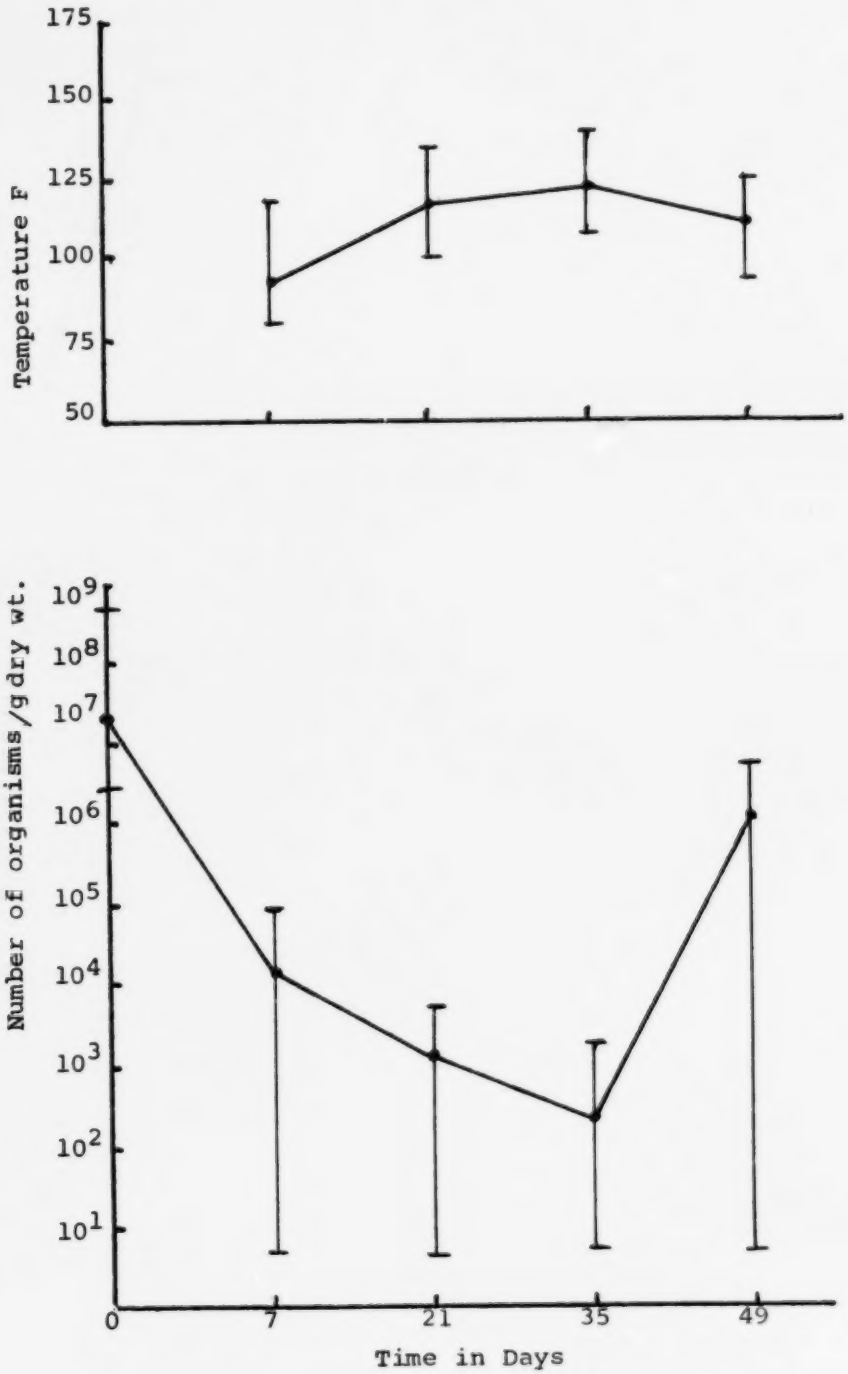


FIG. 8. Average total coliform •MPN from eight refuse-sewage sludge windrows. Vertical bars represent extreme variations found in the windrows. 2 in. from bottom.

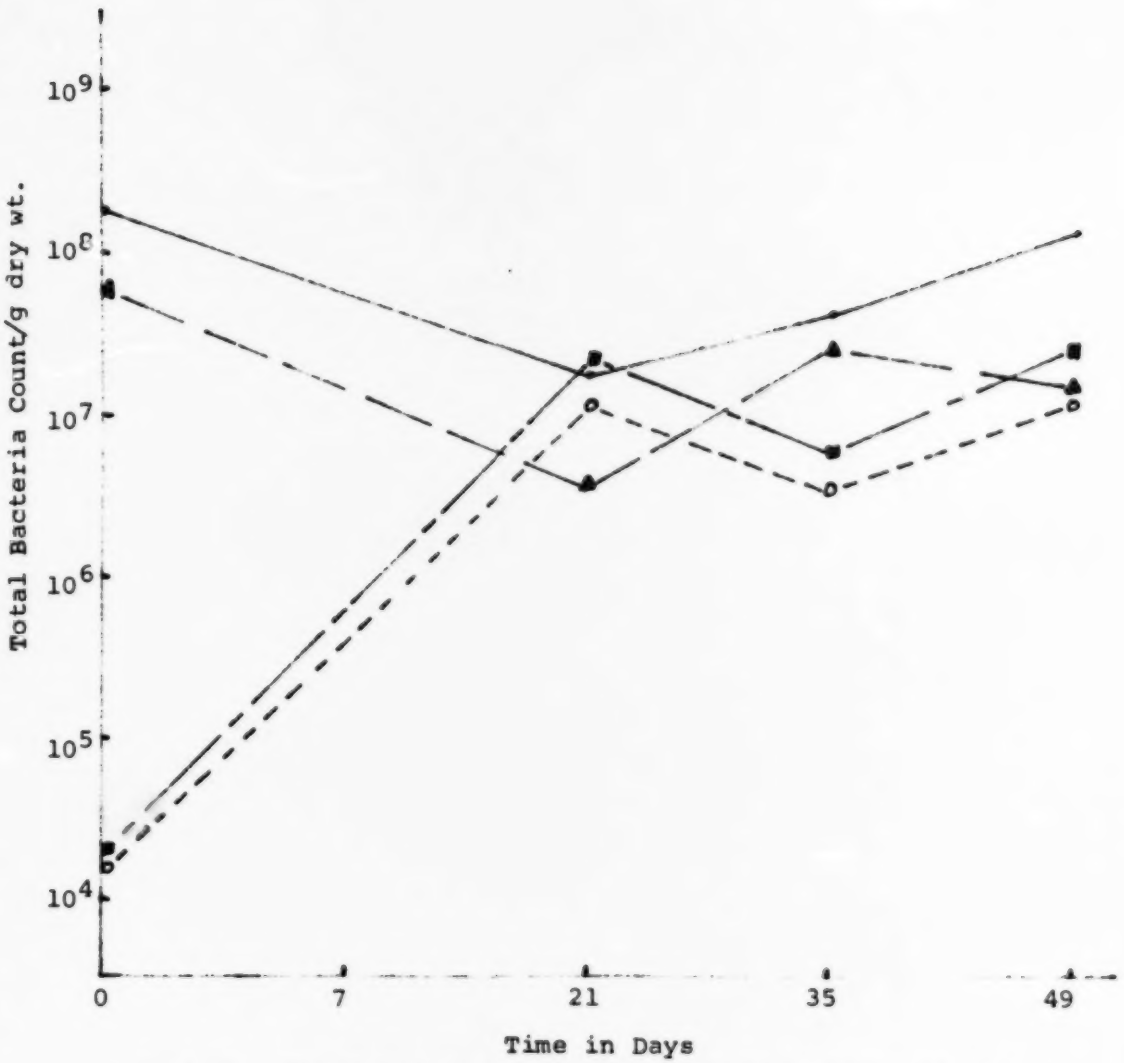


FIG. 9. Total plate count of raw refuse windrow 11-8-67 - 12-27-67. Each plate count was made from composites of 8 samples. Plates prepared in duplicate and incubated at 95F and 131F under aerobic and anaerobic conditions. 95F aerobic ●——●; 95F anaerobic ▲— — —▲; 131F aerobic ■——-——■; 131F anaerobic ●- - - - -○.

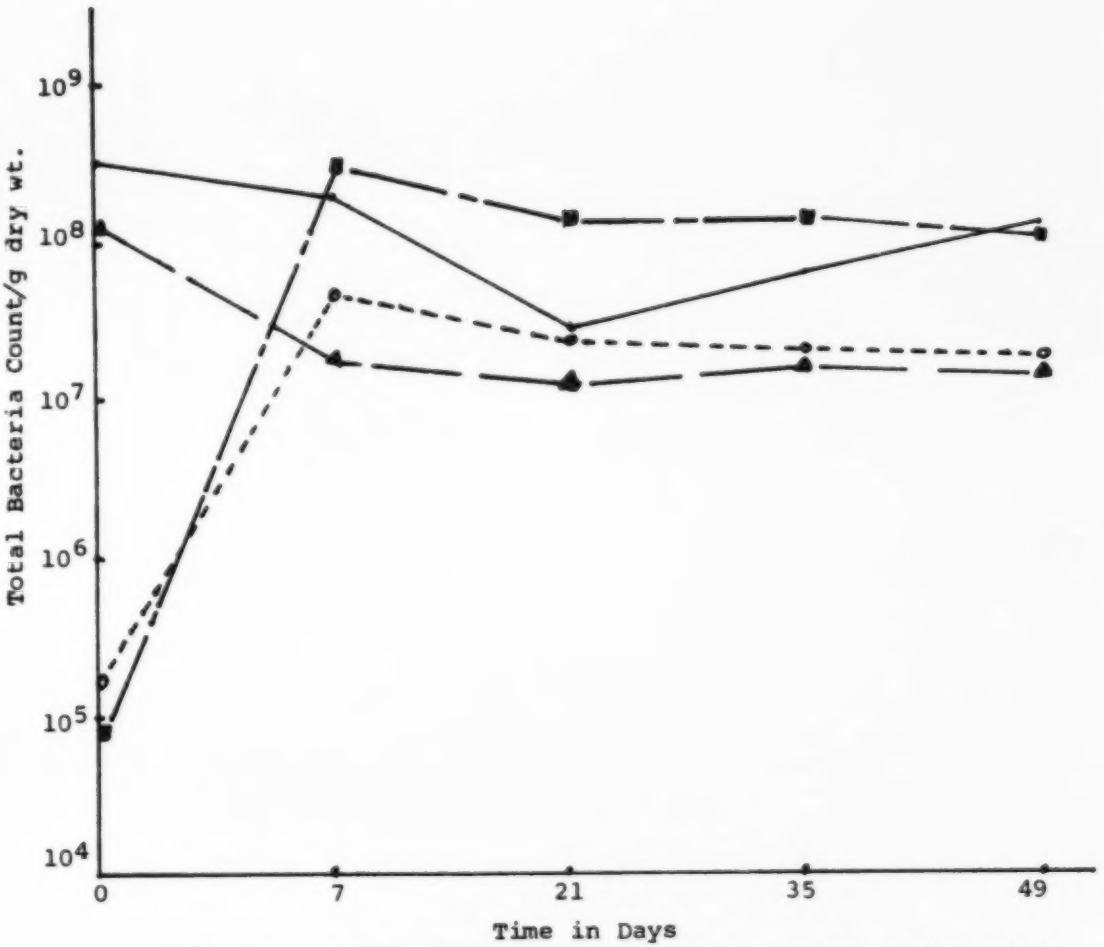


FIG. 10. Total plate counts of refuse-sewage sludge windrow. The refuse-sewage sludge windrow plate counts represents the average counts obtained from three windrows. All plates were prepared in duplicate and incubated at 95F or 131F under aerobic and anaerobic conditions. 95F aerobic ——— ; 95F anaerobic ▲— —▲ ; 131F aerobic ■— – —■ ; 131F anaerobic ●— – – –● .

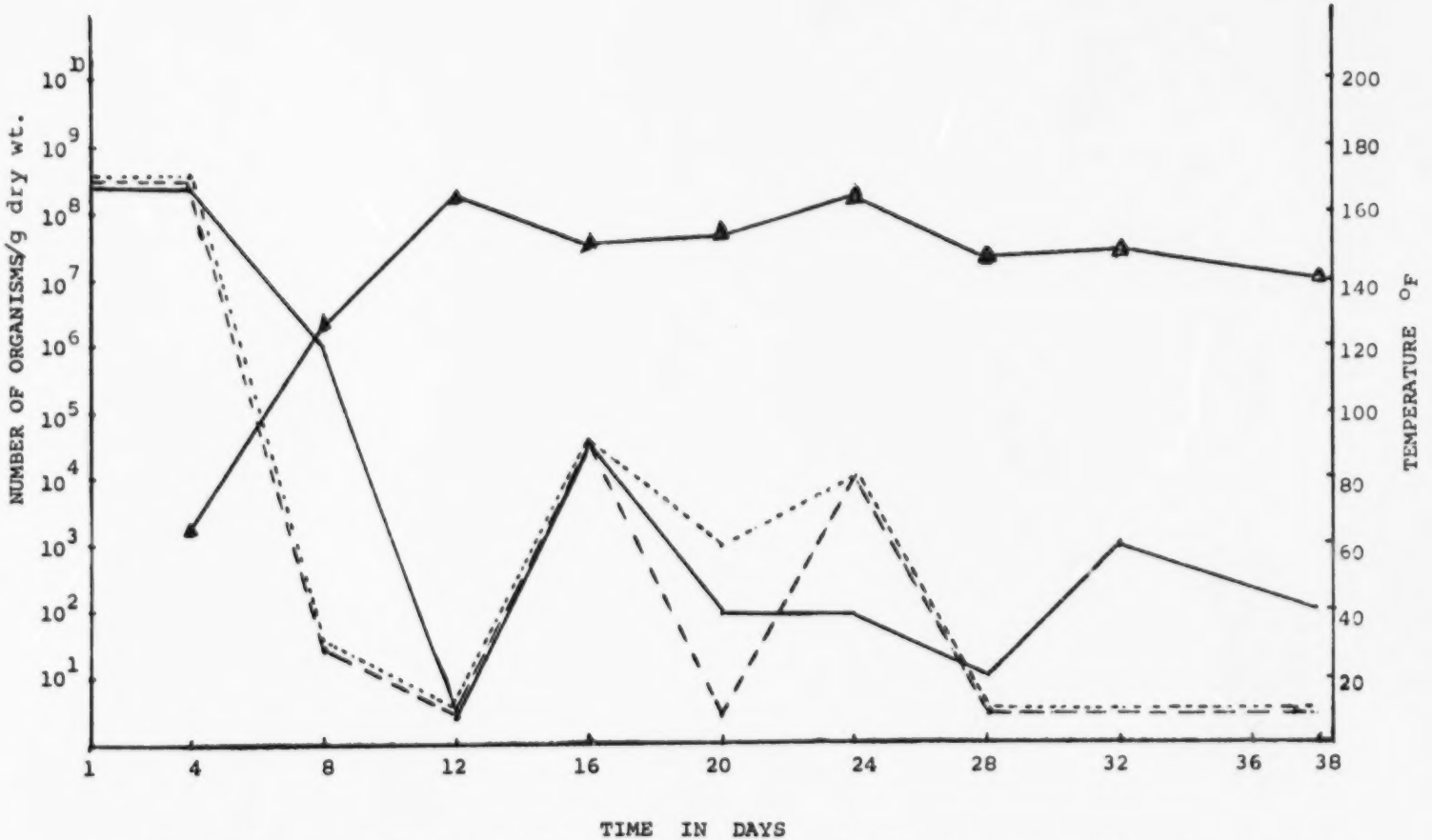


FIG. 11. Survival time in a typical windrow (13-J) of total coliforms - - - - - -; fecal coliforms — — — —; and fecal streptococci ———— . Temperature ▲———▲ .

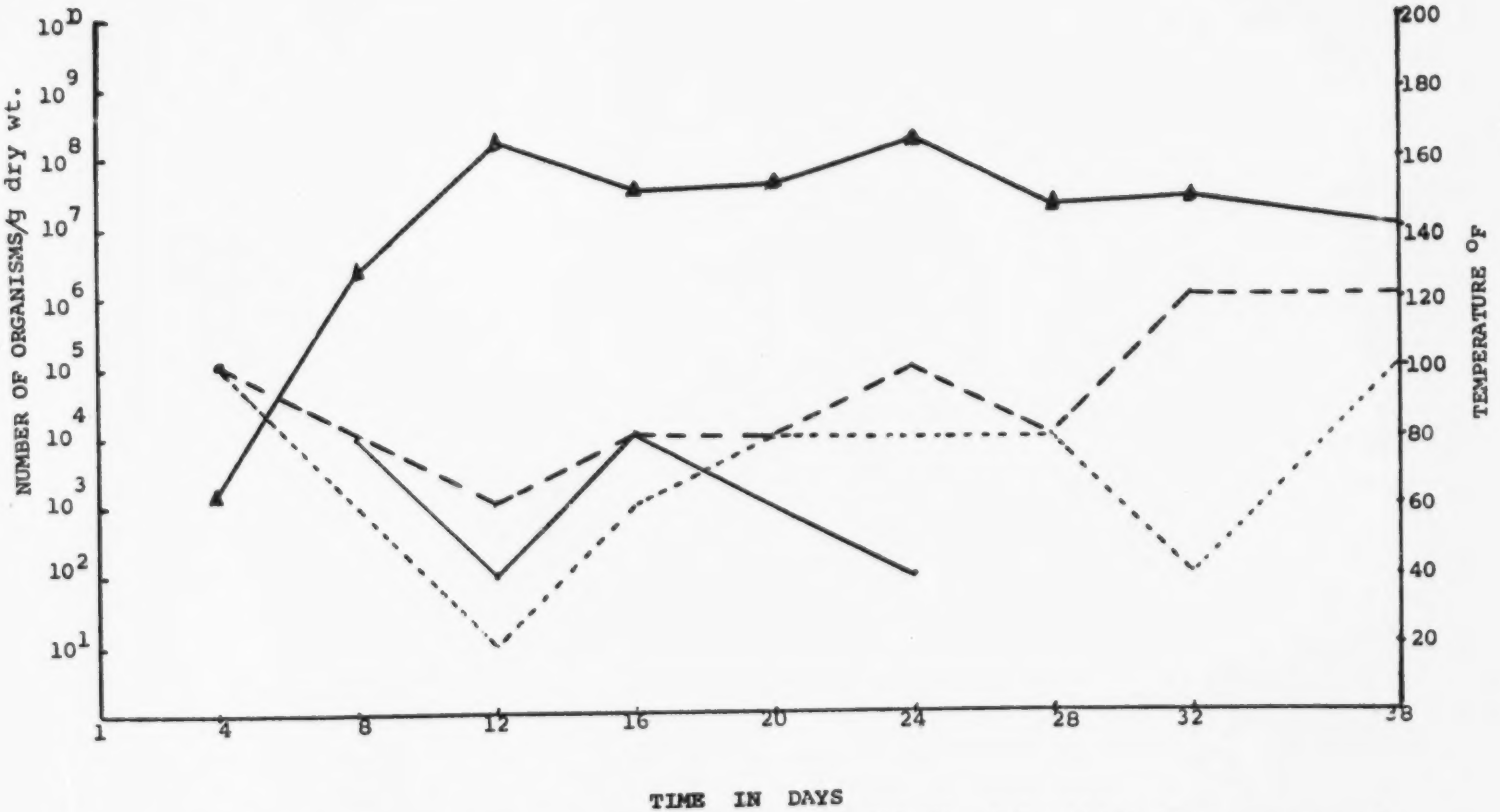


FIG. 12. Survival time in a typical windrow (13-J) of Bacillus sp.— — — —; Chromogenic sp. - - - - - - - -; and pigmented Pseudomonads ———— . Temperature ▲———▲

chromogenic colonies reappeared in significant numbers in the compost and remained evident for the duration of the process. These organisms were found to be gram positive to gram variable, spore forming pseudothermophilic or thermophilic Bacillus. Typical blue-green pigmented Pseudomonas colonies were found in the compost after 4 to 7 days but disappeared in 18 to 24 days.

It was of interest to find that species of Proteus and coagulase-positive staphylococci could be isolated in large numbers from raw refuse but were never found in raw sewage sludge. These organisms disappeared rapidly, however, as the compost temperature increases (FIG. 13). Eighty per cent of the Proteus strains isolated were found to be P. mirabilis, P. vulgaris was never isolated from any samples examined.

The presence of Proteus in raw refuse and its disappearance from compost, closely paralleling the disappearance of pathogenic enteric bacilli inserted in the windrows, suggests that this bacillus could be utilized as an indicator organism of raw refuse.

Total coliform an fecal coliforms were isolated from all of the soil samples examined whereas Proteus was present only in those soil samples containing decaying matter (Table 2). When raw refuse was added to garden soil Proteus could be isolated from the area only as long as the refuse remained. When the refuse decomposed and disappeared from the soil, Proteus could no longer be recovered.

Parasites have been observed, by direct and concentrated wet mounts preparations, periodically from concentrated sewage and refuse-sewage sludge compost. The parasite ova most frequently observed were those of Ascaris, hookworm, Trichuris, Trichostrongylus sp. and H. diminuta. Forty-two percent of raw dewatered sewage and first stage sludge samples examined contained at least one parasite protozoan ova or cyst. Parasites were observed in 33% of the finished (49th day) compost samples. None of the samples were heavily infested, most contained only one parasite. Of the total number of samples examined, parsites were observed in only 5-8% of the specimens. The parasites observed in the finished product were characteristic of those associated with bird and animal infestations and

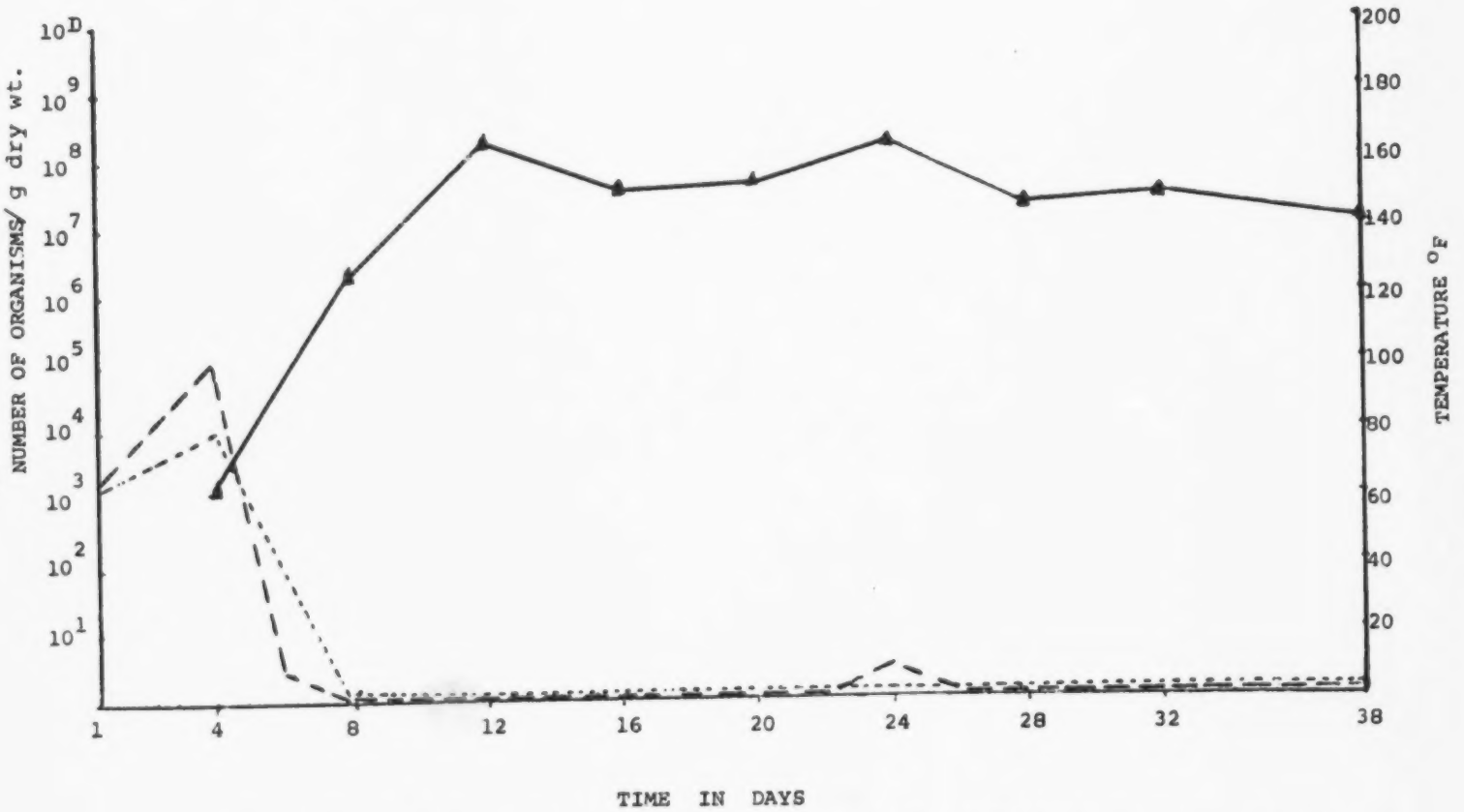


FIG. 13. Survival time in a typical windrow (13-J) of Proteus — — — ; and coagulase-positive staphylococci- - - - - - - - - . Temperature ▲——▲

TABLE 2. *Proteus* and coliforms in soil

Soil Sample	Coliforms*		Proteus*
	Total MPN	Fecal MPN	
Pasture	6.6×10^5	9.8×10^3	Negative
Garden	2.2×10^5	6.6×10^2	Negative
Woodland	1.6×10^3	4.1×10^1	Positive

*Per 5 grams soil sample

were not of human origin.

During the course of this investigation enteroviruses were never isolated from sewage sludge, raw refuse or refuse-sewage sludge mixtures. Subsequent investigation revealed that no enteroviruses had been isolated by the Tennessee Department of Public Health Department from human fecal specimens submitted from the Upper East Tennessee area from April 1964 to July 1, 1968. A large number of Echo 9 viruses were isolated in 1958 and 1959 (6). It is not unexpected, therefore, that these viruses could not be isolated from compost.

Pathogenic fungi could not be demonstrated in any of the specimens of refuse-sewage sludge compost submitted to the Tennessee Department of Public Health Laboratories indicating either their absence in compost or their presence only in extremely small numbers.

The results obtained from the insertion studies confirm previous findings that the bacterial population of the windrow is directly related to its internal temperature. We were unable to demonstrate any type of antagonistic action resulting from antibiotic activity or other metabolic products of microorganisms in compost. Aqueous, ether, chloroform, alcohol, benzene or combined solvent extracts of large quantities of compost, at various stages in the process, were neither bactericidal nor bacteriostatic for a wide variety of gram positive and gram negative bacteria.

The gram negative pathogenic enteric bacilli do not survive the normal windrow composting environment. Species of Salmonella or Shigella originally present in refuse or sewage or inserted under controlled conditions into the refuse-sewage sludge windrows disappeared from the windrows within 7 to 21 days. Figure 14 shows the survival of E. coli, S. aureus, S. typhimurium, and Sh. sonnei inserted in a windrow in January, 1969 when the temperature of the refuse-sewage sludge mixture was approximately 25 F (-4 C). The temperature of the windrow did not reach 120 F (49 C) until about 21 days. Under these conditions the cultures not only survived but actually increased in number through the 14th day. As the temperature approached 120 F the bacterial population declined. While the Salmonella and Shigella species were completely eliminated by the 28th

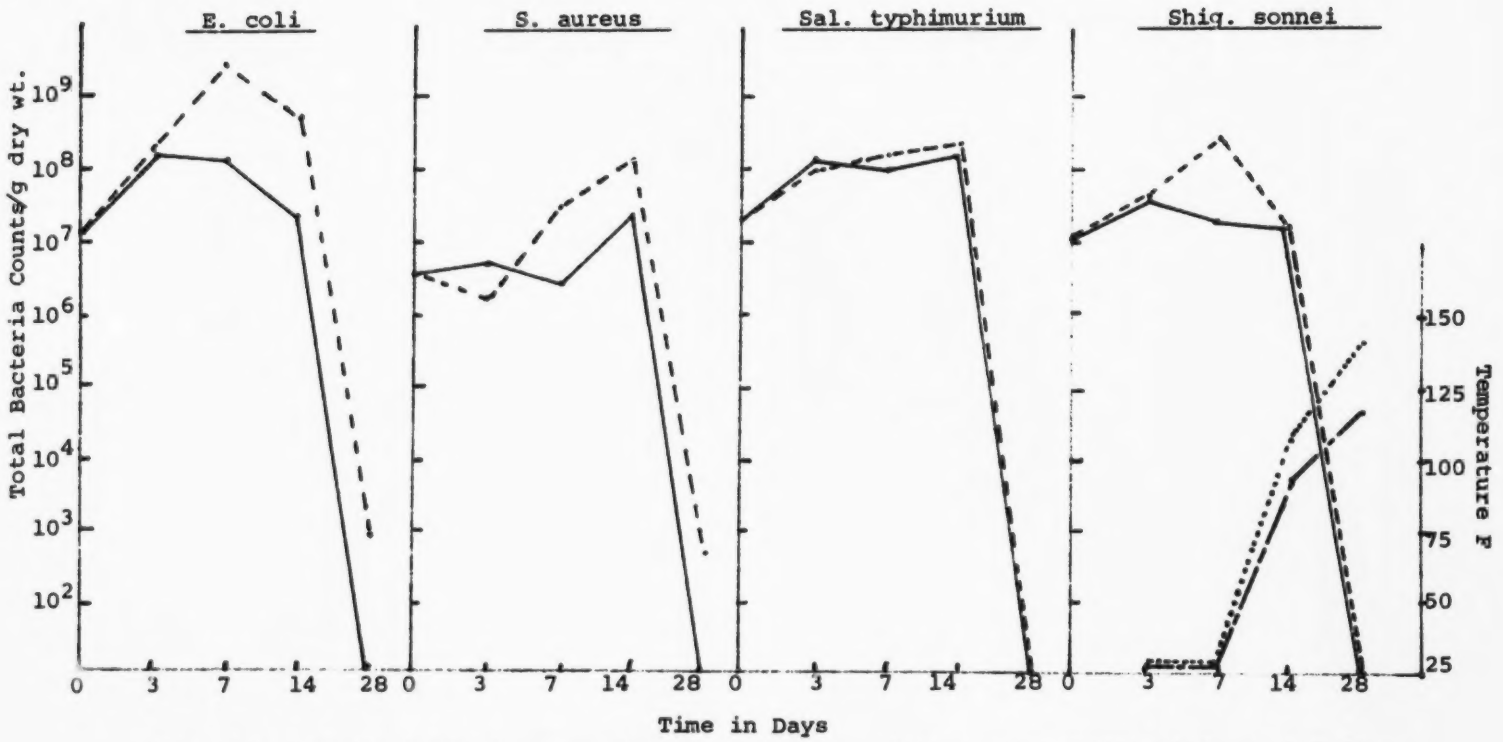


FIG. 14. Survival of microorganisms inserted in refuse-sewage sludge windrow. 1-7-69 to 2-4-69. Sealed culture vials inserted in windrow at 2 in. and mid-depth. Vials were removed on days indicated and viable plate counts made of surviving bacteria. 2 in. depth — — — — mid-depth _____; 2 in. temperature ..... ; mid-depth temperature ——-——.

day, E. coli and S. aureus cells were still viable at the 2 in depth on the 28th day. The temperature at the 2 in. depth did not reach 120 F until approximately the 28th day. Figure 15 illustrates the survival of these same microorganisms inserted in a windrow on a warm September day when the temperature of the windrow was slightly over 100 F (38 C) by the time the cultures were inserted. All the microorganisms were completely eliminated by the 4th day when the temperature rapidly reached 140 F (60 C). The results of survival studies carried out with the four bacteria in sealed tubes and on saturated filter paper discs are shown in Table 3. Also shown are the results obtained from the insertion of Geotrichum candidum and Aspergillus fumigatus. It is obvious from these results that despite the somewhat erratic temperature patterns of the top 2 to 4 in. and the bottom 2 to 4 in. sections of the windrow, all bacteria were killed by the 27th day, and the fungi by the 35th day. In all of these studies the microorganisms inserted in windrows which followed a normal temperature pattern were rapidly killed. The results in Table 4 are included to examplify the extreme variation observed in one of the windrows. The temperature of this windrow was erratic and as a result E. coli, S. typhimurium and Sh. sonnei survived through the 24th day. Poliovirus was inactivated by the 7th day. One culture of Geotrichum candidum survived 24 days and Aspergillus fumigatus, inserted after the windrow was 14 days old, was killed within 10 days. These results are of interest because this windrow was perhaps the most erratic of all the windrows studied. However noe of the organisms survived 27 days in spite of the irregularities.

Leptospira philadelphia did not survive the windrow temperature for more than 2 days. Cultures of Blastomyces dermatidis and Histoplasma capsulatum generally did not survive a 7 day exposure to the windrow as illustrated in Table 5.

Cysts of Endolimax nana and Entamoeba histolytica, as well as hookworm ova (Necator americanus or Ancylostoma duodenale) obtained as stool specimens from hospital patients disappeared (disintergrated) after 7 days exposure to the windrow environment (Table 6). On the other hand, as shown in Table 7, hookworm, tapeworm and whipworm ova from dog feces were observed in specimens 35 days after

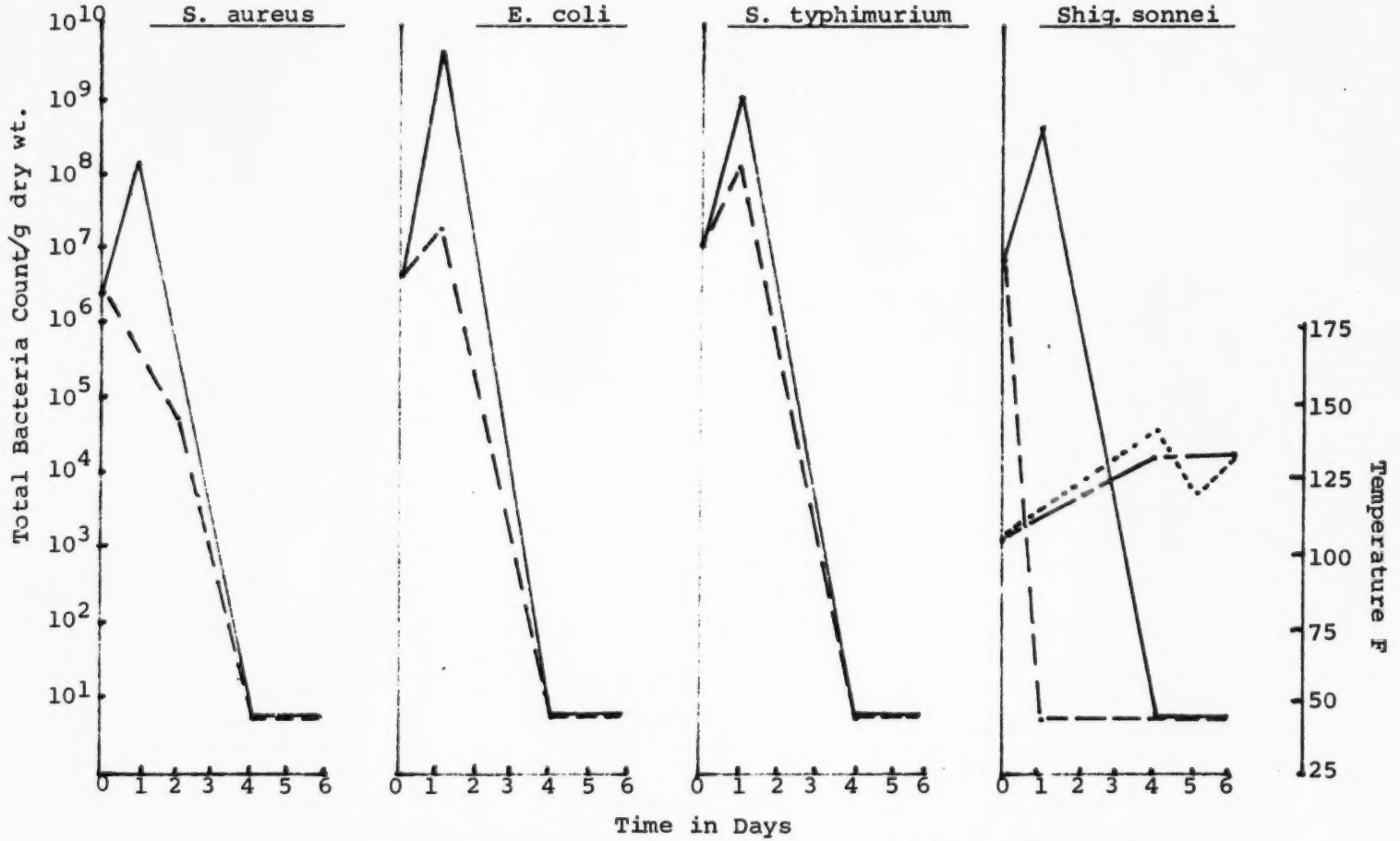


FIG. 15. Survival of microorganisms inserted in refuse-sewage sludge windrow 9-5-68 to 9-12-68. Sealed culture vials inserted in windrow at 2 in. and mid-depth. Vials were removed on days indicated and viable plate counts made on surviving bacteria. 2 in. depth - - - - - - - - ; mid-depth ———————— · 2 in. temperature ·····; mid-depth temperature——— - ——— - ——— .

TABLE 3. Survival of microorganisms inserted in refuse-sewage sludge windrow 1-23-69 to 2-27-69

Day	Depth	Temp. F.	E. coli		S. aureus		S. typhimurium		Sh. sonnei		G. candidum	A. fumigatus
			Tube	Disc	Tube	Disc	Tube	Disc	Tube	Disc	Tube	Tube
0			4.2×10^7	+	4.5×10^6	+	7.5×10^7	+	3.2×10^7	+	+	+
7	2"	102	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	x	+
	Mid	122	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	+	+
	Bot.	136	$< 10^1$	-	$< 10^1$	-	2.2×10^4	-	$< 10^1$	-	+	+
14	2"	88	$< 10^1$	+	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	+	-
	Mid	110	4.5×10^2	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	+	+
	Bot.	96	$< 10^1$	-	$< 10^1$		2.3×10^7	-	1.0×10^2	-	-	+
27	2"	144	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	+	-
	Mid	152	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	-	-
	Bot.	96	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	-	-
35	2"	122	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	x	-
	Mid	142	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	-	-
	Bot.	98	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	$< 10^1$	-	-	-

- = No growth; + = growth; x = contaminated or broken tube

TABLE 4. Survival of microorganisms inserted in refuse-sewage sludge windrow 1-17-68 to 2-13-69

Day	Depth	F	E. coli	S. aureus	S. typhimurium	Sh. sonnei	Polio Virus Type II	G. candidum	A. fumigatus
0			7.3×10^7	2.9×10^7	4.2×10^7	3.5×10^7	3000 TCID50	+	Inserted after windrow was 14 days old
7	2"	110	1.3×10^8	4.8×10^7	5.0×10^6	3.1×10^3	—	X	
	Mid	120	1.6×10^6	1.8×10^5	$< 10^1$	$< 10^1$	—	+	
	Bot.	140	$< 10^1$	8.5×10^2	$< 10^1$	1.6×10^2	—	+	
14	2"	124	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	X	+
	Mid	148	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	X	+
	Bot.	140	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	X	+
24	2"	70	3.5×10^4	$< 10^1$	5.0×10^2	1.0×10^2	—	+	—
	Mid	118	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	—	—
	Bot.	64	7.5×10^2	$< 10^1$	$< 10^1$	5.0×10^2	—	—	—
27	2"	146	$< 10^1$	$< 10^1$	X	$< 10^1$	—	—	—
	Mid	148	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	—	—
	Bot.	102	$< 10^1$	$< 10^1$	$< 10^1$	$< 10^1$	—	X	—

___ = No growth; + = growth; X = contaminated or broken tube. Discs saturated with cultures of the bacteria listed in the Table were negative by the 7th day.

TABLE 5. Survival of microorganisms inserted in refuse-sewage sludge windrow 10-21-68 to 11-12-68

Day	Depth	Temp. F	L. philadelphia	B. dermatidis	H. capsulatum
0		110	+	+	+
2	2"	96	-	Specimens taken only from 7th to 22nd days	Specimens taken only from 7th to 22nd days
	Mid	138			
3	2"	124	-		
	Mid	114	-		
4	2"	120	-		
	Mid	-	X		
7	2"	124	-	-	-
	Mid	144	-	-	-
9	2"	140	X	-	-
	Mid	154	-	-	-
14	2"	142	-	-	-
	Mid	158	-	-	-
22	2"	128	-	-	-
	Mid	152	-	-	-

+ = growth; - = no growth; X = broken or lost in windrow

TABLE 6. Survival of human parasites inserted in refuse-sewage sludge windrows 9-5-68 to 10-2-68

Day	Depth	Temp. F	Cysts		Hookworm ova
			E. nana	E. histo-lytica	
0			+++	++	+++
7	Midpoint	140	-	-	-
14	Midpoint	153	-	-	-
21	Midpoint	159	-	-	-
28	Midpoint	141	-	-	-

+++ = Heavy infestation-many cysts or ova observed in wet mount; ++ = several cysts or ova observed in wet mount; - = no cysts or ova observed in wet mount.

TABLE 7. Survival of dog parasites inserted in refuse-sewage sludge windrows 12-11-68 - 1-20-69

Day	Depth	Temp. F	Ova		
			Hookworm	Tapeworm	Trichurius
0			+++	+++	+++
6	2"	137.5	+++	+++	+++
	Mid	--	+++	+++	X
20	2"	159	X	+++	+++
	Mid	--	+++	+++	+++
27	2"	120	+++	+++	+++
	Mid	150	+++	+++	X
35	2"	124	+++	+++	+++
	Mid	--	X	X	X

+++ = Heavy infestation; large numbers of intact ova observed in wet mounts.

being inserted into windrows. While these ova were present in large numbers it was not possible to determine their viability. It would appear from these results that parasitic ova and cysts from human infections are more susceptible to disintegration by the composting process than are dog parasitic ova. Although parasitic ova may remain intact they may no longer be viable. Unfortunately funds were not available to complete this phase of the study.

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TECHNICAL REPORT DATA
(Please read Instructions on the reverse before completing)

1. REPORT NO.	2.	3. RECIPIENT'S ACCESSION NO.
EPA-670/2-75-023		

4. TITLE AND SUBTITLE	5. REPORT DATE
EVALUATION OF HEALTH HAZARDS ASSOCIATED WITH SOLID WASTE/SEWAGE SLUDGE MIXTURES	April 1975; Issuing Date
	6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S)	8. PERFORMING ORGANIZATION REPORT NO.
William L. Gaby	
9. PERFORMING ORGANIZATION NAME AND ADDRESS	10. PROGRAM ELEMENT NO.
Department of Health Sciences East Tennessee State University Johnson City, Tennessee	1DB064;ROAP 24ALU;Task 03
	11. CONTRACT/GRANT NO.
	68-03-0128
12. SPONSORING AGENCY NAME AND ADDRESS	13. TYPE OF REPORT AND PERIOD COVERED
National Environmental Research Center Office of Research and Development U.S. Environmental Protection Agency Cincinnati, Ohio 45268	Final
	14. SPONSORING AGENCY CODE

15. SUPPLEMENTARY NOTES

Contact - Clarence A. Clemons 513/684-4481

16. ABSTRACT

This report summarizes and evaluates the health hazards associated with municipal solid waste/sewage sludge composting by the windrow composting process. The occurrence and survival of pathogens, parasites, and indicator bacteria at various stages during the composting process are described. The study shows that windrow temperatures of 120F to 167F (49C-74C) maintained for at least 7 days destroy pathogens and human parasites. Dog parasitic ova, however, remain intact 35 days after exposure. Considerable variation in the temperature is found at the top and bottom 2 inches of the windrow indicating that proper turning of the compost is essential to ensure destruction of pathogens and parasites. It is concluded that a properly composted solid waste or solid waste/sewage sludge mixture is microbiologically acceptable as a soil conditioner for gardens, farms, and lawns, or for filling areas of erosion without creating health hazards.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Composting Survival Parasites Sewage Sludge	Solid waste Sewage sludge Pathogens Health aspects	13B

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS *(This Report)*	21. NO. OF PAGES
RELEASE TO PUBLIC	UNCLASSIFIED	56
	20. SECURITY CLASS *(This page)*	22. PRICE
	UNCLASSIFIED	

EPA Form 2220-1 (9-73)

U.S. GOVERNMENT PRINTING OFFICE: 1975-657-592/5359 Region No. 5-11